

**Assessment of growth performance, haemo-
biochemistry and meat quality of Japanese
quails fed on *Moringa oleifera***

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Dissertation accepted in fulfilment of the requirements for
the degree *Masters of Science in Animal Science* at the
North West University

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DECLARATION

I declare that this dissertation submitted to the North-West University for the degree of Master of Science in Agriculture in Animal Science has not been previously submitted to any other university or institution and that it is my own original work. Material and information from other sources are fully recognized and acknowledged.

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GENERAL ABSTRACT

The use of readily available, non-conventional feed resources in quail farming has the potential to sustainably improve animal protein supply in resource-poor communities in South Africa. *Moringa oleifera* leaf meal is one such feed resource that is rich in potentially beneficial bioactive compounds with nutraceutical properties but its utility has not been assessed in quails. This study was, therefore, conducted to determine the effect of *M. oleifera* leaf meal (MOLM) on growth performance, haematological and serum biochemical indices, carcass characteristics and meat quality of Japanese quails. A total of 400, one-week old, mixed gender Japanese quails (196.6 ± 3.12 g body weight) were reared using a commercial starter diet and had access to fresh water while ambient temperature was maintained via infrared lamps until 4 weeks of age. At five weeks of age, 224 female Japanese quails were obtained upon gender sexing. The female quails were then randomly and evenly allocated to 28 pens (experimental units) with each pen holding eight quails. The MOLM was used to dilute a commercial grower diet at a rate of 0, 25, 50 and 75 g/kg (CON, MOLM25, MOLM50 and MOLM75, respectively), producing four isonitrogenous and isoenergetic dietary treatments. The four diets were randomly allocated to the 28 pens resulting in seven replicates per dietary treatment. During the feeding trial feed intake and body weight gain were measured and used to calculate feed conversion efficiency (FCE). At 10 weeks of age, all quails were slaughtered at a local abattoir. Blood was collected during slaughter for analyses of haematological and serum biochemical indices. Repeated measures analysis showed no significant week $\times$ diet interaction effect on weekly feed intake, weekly body weight gain and weekly FCE. Dietary treatments had a significant effect on overall body weight gain ($P < 0.05$). Japanese quails fed MOLM25 had higher ($P < 0.05$) body weight gain (30.72 g/bird) compared to those fed on MOLM50 (18.60 g/bird). Quails offered MOLM25 had higher haemoglobin (14.85 g/dL) compared to those fed diets MOLM75 (12.69 g/dL), MOLM50 (13.70 g/dL) and CON (14.32 g/dL). There were

dietary effects ($P < 0.05$) on the length of large intestine and weight of gizzards. Japanese quails offered MOLM75 (14.13 cm) had the longer large intestines compared to Japanese quails offered MOLM50 (11.24 cm), CON (12.73 cm) and MOLM25 (13.84 cm), and, similarly, the highest (5.45 g) gizzard weight was observed in quails fed MOLM75. It is, therefore, recommended that the inclusion of *Moringa oleifera* leaf meal in Japanese quails diets be at optimum level of 25 g/kg.

Keywords: Blood indices; feed additives; growth; Japanese quails; *Moringa oleifera* leaf meal.

ACKNOWLEDGEMENTS

Firstly, I acknowledge the Almighty God for the gift of life and for the strength he bestowed on me to be able to complete the requirements for this degree. My special gratitude goes to my supervisors, Dr C.M. Mnisi and Prof. V. Mlambo, who guided and motivated me during this study. I would not have persevered and completed this study had they not supported and pushed me. I am really grateful to my colleagues: Mr L.T. Nhlane and Mr W. Mankga, as well as Animal Science staff members: Dr K.E. Ravhuhali and Miss C. Kumanda, who willingly helped during the experimental work. Special thanks also go to my family for the moral support and love they gave me.

DEDICATION

This work is dedicated to my family, who have been financially and morally supportive throughout my study. My parents Mr and Mrs Mulaudzi, my siblings Thendo, Zwiswa, Lisani, Ngaahule, Ankonisaho and not forgetting my friend Rambasa Edzani. Thank you for encouraging me, all this is for you.

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LIST OF ABBREVIATIONS

AAs	: Amino acids
AWFI	: Average weekly feed intake
AGPs	: Antibiotic growth promoters
ALP	: Alkaline phosphatase
ALT	: Alanine aminotransferase
AWFI	: Average weekly feed intake
AWG	: Average weight gain
CCW	: Cold carcass weight
CP	: Crude protein
DM	: Dry matter
FCE	: Feed conversion efficiency
FI	: Feed intake
GLM	: General linear model
HCW	: Hot carcass weight
LSMEANS	: Least square means
MOLM	: <i>Moringa oleifera</i> leaf meal
NRC	: National Research Council:
OM	: Organic matter
SBM	: Soybean meal
SEM	: Standard error of the mean.
WHC	: Water holding capacity

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1 CHAPTER ONE - INTRODUCTION

1.1 Background

Poultry production is one of the quickest ways through which the growing demand for animal proteins in Africa can be met. This is because chickens have short generation intervals, high rate of reproduction and can efficiently transform nutrients into high quality protein (Akinfela *et al.*, 1999). Currently, poultry meat and eggs are by far the most consumed (FAO, 2012), accounting for more than 30% of all animal protein consumption (Permin & Pedersen, 2000). There is, however, a need to diversify the type of poultry species available for commercial production given the disease and other production challenges that face traditional avian species. One potential bird that could revolutionize the poultry industry in developing, resource-poor countries is the quail. Quail farming is a new enterprise in the South African poultry industry (Mnisi & Mlambo, 2018). However, if sufficiently and sustainably developed, it can contribute immensely to animal protein supply, enabling the poultry industry to guarantee food and nutrition security as well as economic growth. Quail meat is high in protein, polyunsaturated fatty acids and essential trace minerals (Mnisi & Mlambo, 2018). Quail eggs can help reduce problems related to kidney and liver or gallbladder in humans. The nutritional value of quail eggs is much higher than that of other poultry eggs because they are richer sources of antioxidants, minerals, and vitamins than other foods (Lalwani, 2011). Besides, quail meat and eggs are tastier than those of chickens and has less fat. Quail products have been shown to promote body and brain development in children and nursing mothers (Sahim *et al.*, 2008). The widely distributed quail species is the *Coturnix coturnix japonica*, commonly known as the Japanese quail. This quail species has also been widely used as a research model for biological and genetic studies (Nasar, *et al.*, 2016) Tsudzuki, 1994; Narayan *et al.*, 1998). It has a small body's (± 160 g) size, it is easy to handle, and a large number of these birds can be

reared in a space required for a single adult chicken. Their sexual maturation is rapidly attained at six weeks of age. Quails are prolific birds producing a large number (300 per year) of eggs thus a large generation of offspring can be produced from a few parents. In recent years, the quail sector has been one of the largest and fastest growing agro-industries throughout the world, however, the cost of poultry feeds has remained a significant challenge. This is because poultry production, especially in the tropics, faces the challenge of high prices of the conventional protein and energy sources. The problem has been worsened due to the increasing competition between humans and livestock for these protein ingredients as food (Nwekirk, 2010). Non-conventional feedstuffs, with no value as direct food resources for humans, need to be identified for use in poultry diets so as to reduce the cost of production. Indeed, leaf meal protein sources are amongst feed sources being investigated in recent times and do not only serve as a source of protein, but provides some necessary vitamins and minerals (Onu & Otuma, 2008) as well as bioactive compounds that have the potential to boost growth performance as well as meat quality. Locally available nutraceutical plants such as *Moringa oleifera*, commonly called horseradish tree or drumstick tree, are potential sources of nutrients and bioactive compounds for Japanese quails. *Moringa oleifera* is widely distributed in many tropical and subtropical regions. It has an impressive range of medicinal uses owing to the antimicrobial and antioxidant properties of its bioactive compounds (Bennett *et al.*, 2003; Ferreira *et al.*, 2008).

1.2 Problem statement

The poultry industry has been a reliable protein source for human beings for many years (Ayasan, 2013). However, production costs have increased due to cost of feed, energy (electricity), vaccines and chemotherapeutic drugs. The use of antibiotics as growth promoters in the poultry industry has been banned in some jurisdictions due to concerns relating to

harmful effects on human health. These harmful effects include the development of microbial resistance to antibiotics and the presence of antibiotic residues in chicken products destined for human consumption (William & Losa, 2001; McCartney, 2002). Consequently, herbs, spices, and various plant extracts, considered to be natural products that consumers would accept, have received increased attention as possible feed additives in place of antibiotic growth promoters (Catalagregori *et al.*, 2008). The growth of human and livestock populations has increased the competition for food and feed between animals and man. Furthermore, the use of feed additives such as vitamin E and selenium, which are usually incorporated in monogastric feeds raise the cost of feeding (Bergh *et al.*, 2012). Medicinal plants have been proposed as natural alternatives to antibiotic growth promoters. Of late, these plant-based remedies have found an application in poultry production as enhancers of growth performance through the action of bioactive secondary plant compounds (Saki *et al.*, 2012). One such plant is *Moringa oleifera* but its effect on growth performance, blood indices, carcass traits and meat quality of Japanese quails is unknown.

1.3 Justification

Moringa oleifera leaves are nutritious and contain vitamins, calcium, irons, phosphorus and protein (Murro *et al.*, 2003). Furthermore, heavy metals such as mercury, arsenic and cadmium, which are potentially toxic are absent from the leaves of *M. oleifera*, thus making their incorporation into poultry diets safe (Donkor *et al.*, 2013). The presence of vitamin C, vitamin E, carotenoids, flavonoids and selenium makes *M. oleifera* a potential antioxidant source (Moyo *et al.*, 2012). The antioxidant compounds (phenols, vitamin C, vitamin E, β -carotene, zinc, selenium, flavonoids) in *M. oleifera* improve shelf-life and the quality of meat products in the pre-slaughter or post-slaughter stages (Valeria & Williams, 2011). *Moringa oleifera* also lowers the activity of pathogenic bacteria and moulds thus helping to protect chickens from

subclinical infections and thus boost their growth performance (Gaia, 2005). Large amounts of high quality forage from *M. oleifera* can be obtained without expensive inputs due to favourable soil and climatic conditions for its growth (Makkar & Becker, 1997). Indeed, Sarwatt *et al.* (2004) reported that *M. oleifera* foliage is a potential inexpensive protein source for livestock feed. This work highlights the growth performance, haemo-biochemistry, carcass and meat quality traits in growing Japanese quails as influenced by dietary moringa leaf meal.

1.4 Objectives

The study was designed to investigate the effect of MOLM on growth performance, haematological, serum biochemical parameters, carcass and meat quality traits of Japanese quails. The specific objectives were to:

- i. To determine the effect of *M. oleifera* leaf meal on growth performance of Japanese quails.
- ii. To determine the effect of *M. oleifera* leaf meal on haematological and serum biochemical indices of Japanese quails.
- iii. To determine the effect of *M. oleifera* leaf meal on carcass characteristics and meat quality measures in Japanese quails.

1.5 Hypotheses

The study tested the alternative hypothesis that inclusion of *M. oleifera* leaf meal improves physiological and meat quality responses of Japanese quails.

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2 CHAPTER TWO – LITERATURE REVIEW

2.1 Introduction

In South Africa, the poultry industry can be divided into two sub-sectors: commercial and traditional poultry production systems (Ayasan, 2013). In the commercial sector, birds are confined to areas with infrastructure for the entire production cycle, which is necessary for the intensive production and marketing of poultry products. According to DAFF (2017), the per capita consumption of poultry meat for 2017 was 38.08 kg and per capita egg consumption was 7.89 kg in 2017. In communal areas, most households depend on the traditional poultry sub-sector for animal protein (Sonaiya, 2001). This sub-sector provides readily available protein in the form of meat and eggs (Gueye, 1998). The predominant poultry species in the rural areas of Africa are indigenous chickens (*Gallus domesticus*) (Andrews, 1990; Jalaludin, 1992). The low productivity of unimproved indigenous chicken strains may be attributed to slow growth rates, prevalence of predators, poor feeding, and disease outbreaks (Nasar, 2016). There is, therefore, a need to introduce additional poultry species with a shorter life cycle to increase animal protein supply by complementing the existing poultry species.

A possible species for the expansion of the poultry industry reared for both meat and egg production in South Africa is the *Coturnix coturnix japonica*, commonly known as the Japanese quail. All types of climatic conditions are appropriate for quail farming that is why in Japan, commercial quail farming has spread immensely (NRC, 1991). Throughout the world, people are now rearing quails for egg and meat production for both household and commercial purposes. Quail rearing is efficient due to their rapid growth and early attainment of sexual maturity (six weeks of age), high reproductive rates (± 300 eggs per year), disease resistance and low feed requirements (35 g/day/bird) (Murakami & Ariki, 1998; Albino & Barreto, 2003). As a result, Japanese quails are becoming the species of interest in some regions of

Africa (Andrews, 1990; Jalaludin, 1992). The profitability of quails, which depends on egg production, egg size, and survivability of chicks is low. This low profitability might be due to the absence of improved quail strains, predation, diseases, poor nutrition, and sub-optimal management strategies (Alemu, 1995; Alemu & Tadelles, 1997). Quail farming is as profitable as that of chicken, duck and turkey farming, and it plays an important role as an alternative source of protein to ensure food and nutrition demand. Not a lot of labour or capital is needed for a quail farming business (Nasar *et al.*, 2016). because a small pen can accommodate a large number of quails. In addition, other poultry birds can be raised together with quails for the production of eggs and meat.

Nevertheless, there are a lot of challenges facing the poultry industry, such as increased demand of high energy and protein feed ingredients, which are quite expensive. As such, efficient utilization of alternative feed resources that cannot be utilised as food for humans must be fully explored. However, feed costs, remain a major challenge in poultry farming during feed formulation due to the utilization of high priced feed ingredients which constitute about 80% of total cost of production (Elagib *et al.*, 2013). The use of antibiotics as growth promoters in poultry industry has been banned because of harmful effects on human health. Consumers currently perceive edible poultry tissues to be contaminated with harmful concentrations of drug residues linked to the development of antibiotic resistant pathogenic bacteria that poses a threat to human health (Donoghue, 2003). Plant derived additives have are known to contain bio-active compounds with effects similar to antibiotic growth promoters in three main areas, i.e., gut micro-flora, antioxidant properties, and liver function without compromising intestinal health and/or the bird's genetic potential (Hernandez *et al.*, 2004). They are also said to have a potential to modify cholesterol metabolism, leading to a product with healthier implications for human consumption (Wallace *et al.*, 2010). One such plant is *Moringa oleifera* whose leaves have been reported to be rich in both essential and sulphur-containing amino acids, vitamins

C, β -carotene and various phenolic compounds, thus qualifying the leaves as a potential animal feed supplement (Bennett *et al.*, 2003; Ferreira *et al.*, 2008; Moyo *et al.*, 2011) with nutraceutical properties. Many scholars have reported that Moringa leaf meal could play an important role as an ingredient in poultry feeds but its inclusion needs further investigation to validate its effect on growth performance and to ensure safety (Sarwatt *et al.*, 2004).

2.2 Quail farming

Quails can be widely regarded as a source of dietary proteins for human consumption, however, it is important to outline the advantages and disadvantages that comes with quail farming. Ali *et al.* (2012) reported that quail farming aims to diversify and strengthen animal protein production in order to close the gap between demand and supply. One advantage that comes with quail farming is an increase in the domestic poultry produce through eggs as well as meat production (Panda & Singh, 1990). . Another benefit of quail farming is profitability, which comes with the fact that quails are tolerant to numerous avian diseases thus omitting the cost of vaccinations and treatments, which in turn favour the increasing demand for organic products produced with minimal use of antibiotics or antimicrobial growth promoters and other related chemicals (Mnisi *et al.*, 2017). Many countries are farming quails mainly for household consumption and up-market sales (Siddique & Mandal, 1996; Ali *et al.*, 2012). The superior qualities of quails over other poultry species include rapid growth rates, early sexual maturity, short gestation periods and generation intervals (Hemid *et al.*, 2010). Almost all types of weather conditions are suitable for starting quail farming business. Quail meat and eggs are tasty and nutritious, and the eggs are more nutritious when compared to other poultry eggs because they contain more protein, phosphorus, iron, vitamin A, B1 and B2 (Jenkins *et al.*, 2011). Quail farming can play a vital role to meet up with the demand of food and nutrition security. Quails can be reared together with other poultry birds for different purposes.

Furthermore, farmers can raise 6 to 7 quails in a same space required for one adults chicken because of their small body size.



Plate 2.1. Male (left) and female (right) Japanese quails

2.2.1 Importance of quails as new entrants in the poultry industry

The poultry industry is largely dominated by commercial broiler and layer production, with a few indigenous chickens and other birds such as ostrich, ducks and turkey. Expansion of the poultry industry is fundamental to maintain continuous supply of meat and eggs for human consumption (Ayasan, 2013). Raising Japanese quail for food can be regarded as another dimension of poultry farming with its beneficial opportunities and challenges. One benefit of rearing quails is that they support and provide flexibility to chicken production through the supply of meat and eggs (FAO, 1987).

Quail farming needs small capital and low labour (Sanka & Mbagha, 2015) thus one does not need to seek massive amount of funds to venture into quails farming. This provides an opportunity to people who are not financial privileged to start their businesses and grow the poultry industry because there will be more industry players and strong competitors. Dingle

(1990) stated that quail meat has less fat and lean rendering it suitable for health cautious patients. This opens the poultry industry for almost all classes of people who consume meat (Tsudzuki, 1994; Narayan *et al.*, 1998). Quail meat is rich in micronutrients and a wide range of vitamins including B complex, folate and vitamin E and K. It is recommended for people with high cholesterol levels and those who want to maintain a low level of cholesterol (Ihejirikamba, 2012). According to Imchen (2013), quail meat is more tender and juicy than chicken and has less lipid content, its health benefits include promoting body and brain development in children. Good nutrition is crucial to reach the health, it is also an essential role to play in efforts to reach SDGs. It contribute to the fight against poverty and reduces mortality. By indirectly strengthening communities and local economies, good nutrition contributes to the achievement of other development objectives which in turn impact upon the SDGs (Smith, 2005).

2.2.2 *Challenges of quail farming*

Many factors such as high feed costs, predation, diseases and parasites negatively affect the farming of quails. Low productivity results due to challenges associated with sub-optimal nutrition, market inconvenience, lack of information on quail production, poor management, egg cracks, infertility and embryonic deaths (Aboul-Ela *et al.*, 1992). Farming quails do not come as easy as one can elaborate, it also comes with choosing the right breed. Often, farmers struggle when making a decision on which breed to rear, this is because some breeds of quails are feed wasting and aggressive causing injuries to other fellow quails and dominating during feeding, which consequently affect performance (Randall *et al.*, 2008). Feed intake, egg production, body weight gain, nutrient digestibility and feed efficiency tend to decrease with hostile conditions in quails. Although disease outbreaks are limited in quail farming than other poultry birds, there can still be a source of infections if biosecurity measures are not in place.

Furthermore, animals of the same breeds sometimes share common pathogens. The market range of quail is very limited compared to chicken products, this is also because quail egg has some popularity in several regions, but the meat is not yet popularized (Siddiqui *et al.*, 1996). As a consequence of narrower market range, farmers lose interest in quail farming (Siddiqui *et al.*, 1996) causing a fallout on production. Many forms of cannibalism occur in quail raised in captivity such as vent, feather, toe, head and nose pecking. Head and nose pecking is the common type of cannibalism among quails, is generally seen only in adult birds (Randall *et al.*, 2008). Cannibal birds peck at the top of the nose where the fleshy portion merges with the beak and the victim may die from blood loss, but in a case where the bird survives the beak would be permanently deformed and as a result the males cannot be used for breeding. Condition that may result in cannibalism include insufficient feeder or drinker space, underfeeding, insufficient nesting space, overcrowding, nutritional imbalances and nutrient deficiencies, strong artificial light, high ambient temperatures and irritation from external parasites (Randall & Bolla, 2008).

2.3 Types of quail breeds

2.3.1 Japanese quails

There are diverse breeds of quails with over a hundred mostly found in Asia and North-America. These breeds are divided into two main groups: Old World quail and New World quail (OMLET, 2004). The Japanese quail, *Coturnix coturnix japonica*, is a species of the Old World quail found in East Asia that belongs to the order *Galliformes* and the family *Phasianidae* (Minvielle, 2004). These birds are about the size of a plump pigeon that is 38-40 cm. Specific properties for *Coturnix coturnix japonica* such as its ability to produce three to four generations every year make it an interesting research centre animal (Wilson *et al.*, 1961; Reese & Reese, 1962). These birds reach maturity in about six weeks of age and the females

start laying eggs around 50 days of age (Randall & Bolla, 2008). The Japanese quail is a dual-purpose breed of domestic quail reared for both eggs and meat. Under favourable environmental conditions they can produce about 300 eggs per year. In a space of one adult chicken, a flock of 8 to 10 quails can occupy the same space which makes the birds inexpensive to maintain. They are fairly round in shape with females characterised by light tan feathers together with black speckling on the throat and upper breast, whereas the males have rusty brown throat and breast feathers (Minvielle, 2004).

2.3.2 *Button quails*

Button quail, also referred to as Bustard quail, is characterised by its round and small body frame which belong to the family *Turnicidae* of the order *Gruiformes* (Archibald & George, 1991). Button quails are dull in colour and are estimated to be 13 to 19 in length. They run through the grass and are capable of weak whirring flight using their short rounded wings (Fain & Houde, 2004). They usually fly individually or in a flock, and sometimes join flocks of true quails. Females are heavier and stronger with more visible pattern than males, in addition males are less vocal (Paton *et al.*, 2003). These birds reach sexual maturity in about six to eight weeks of age and start laying eggs, and they are capable of laying 200 to 250 eggs a year. They hatch the laid eggs within an incubation period of 12 or 13 days and is the shortest among precocial birds. This quail has strong mothering abilities and care for their chicks with most chicks being independent as early as two weeks, and undergo two speedy moults becoming full sized in six or seven weeks (Archibald & George, 1991).

2.3.3 *Bobwhite quails*

Bobwhite quail (*Colinus virginianus*) originates from the United States, Mexico and the Caribbean. It is a ground-dwelling bird and also a member of the New World quails, scientifically known as *Odontophoridae* (Guthery, 1997). It is the most common species of

quail and often referred to as the number one game-bird of the eastern and southern United States. The name "bobwhite" derives from its characteristic whistling call, which makes the males and females to be easily distinguishable from one another (Brennan, 2007). The males have a white throat and brow stripe bordered by a black colour, whereas the females are similar and have a buff throat and brow without the black border. The overall plumage has grey mottling on the wings, white scalloped stripes on the flanks, and black scallops on the whitish underparts. Both males and females have grey tail with pale legs and feet (Brooks, 2007). The Northern bobwhite quail can range from 24 to 28 cm in length with an average wingspan of 33 to 38 cm (Rollins & Carroll, 2001). This quail is raised mainly as a pet or as a hobby and reach sexual maturity at 16 weeks of age (Butler *et al.*, 2012). The females can lay about 12 to 16 eggs, and a pair may produce one or two broods annually. Both males and females hatch within 23 to 24 days and chicks leave the nest immediately after hatching.

2.3.4 *Gambel quail*

The *Callipepla gambelii* quail, commonly known as the gambel, a name derived from its species, is a member of the New World quail and easily recognized by their top knots and scaly plumage on the undersides. They have a bluish-grey plumage on much of their bodies with an average length of 28 cm and a wingspan of 36 to 41 cm (Matthew, 2001). They weigh 160 and 200 g body weight and is mostly used and raised as a pet (Zink & Blackwell, 1998). Males have a dark and thick plume, black face and neck, and also a black patch on the breast, whereas the females have dull and thin plumes and lack the black markings. The females reach sexual maturity at one year of age and generally lay 10 to 12 eggs, which hatch between 21 and 23 days (Brown, 1998).

2.3.5 Mountain quails

The mountain quail, *Oreortyx pictus*, is a member of the New World quail family. (Zink & Blackwell, 1998). Mountain quail moves fast through brushes and undergrowth. The bird's average length is 26-28 cm with a wingspan of 33-40 cm (Beck *et al.*, 2005). These birds have relatively short, rounded wings and long featherless legs. Females lay between 9 and 10 eggs in a self-made simple scrape concealed in vegetation, usually at the base of trees or shrubs. They reach sexual maturity at 7 to 9 weeks of age. They hatch from 21 to 25 days (Brennan, 1991).

2.3.6 Blue-Scale quails

The Blue-Scale quail also known as *Callipepla squamata*, is a unique and beautiful species of quail. They are referred to as "Cotton Tops" due to the feather arrangements on their heads. They are popular game-birds that range from 10 to 12 inches long at maturity (Sibley and Monroe, 1990). They have grey-brown heads with white "cotton top" crest, black beak, and blue feathers mixed with black from the neck down to the breast and back (Paton & Baker, 2006). From the wings' lower back to the tail feathers, both males and females have a grey-brown colour and as a result it becomes difficult to differentiate them. However, the male's throat is ash-grey while the female's throat is a light yellow (Fain & Houde, 2004), which can be helpful to physically identify the gender. They reach sexual maturity at 12 weeks of age and their breeding season is spring, laying a clutch of 12 to 14 eggs in a ground covered with grasses and other vegetation. Their egg shell is light cream with brown flecks and hatch from 21 to 23 days of laying.

2.4 Nutritional requirements of quails

Nutrition is one of the factors that affects growth and development of an animal. Nutrient requirements vary according to age, stage and purpose (whether the quails are kept for meat or egg) of production. Smith (2005) reported that meeting the dietary requirements for essential nutrients is important to achieve the desired results from the birds. Chinrasri (2004) and Sahin *et al.* (2002) defined nutrient requirements as the amount of nutrients needed to improve the laying ability and hatchability of female animals and to maintain their performance, maximize growth and feed utilization efficiency and also optimize their body fat accumulation. Although all nutrients are important, water is essential (NRC, 1991). However, water intake might be affected by several factors such as temperature, humidity, salt, dietary protein, stage of production, and its ability to resorb water in the kidney (NRC, 1994). Thus quails should have access to clean fresh drinking water at all times because according to Farrell *et al.* (1982), the lack of water for more than 36 h may lead to mortality in quails of all ages. This is because water is important for the transportation of other nutrients like carbohydrates, lipids and protein and also to regulate the body temperature.

Hyankova *et al.* (1997) reported that Japanese quails perform optimally when offered diets containing 216 g/kg crude protein (CP) from day 1 to 35, but the quail's nutrient requirements decline with age as in other birds. For layers, Pinto *et al.* (1998) suggested that the crude protein should be 224 g/kg and is higher than the 18 g/kg suggested by Murakami *et al.* (1993). However, the game-bird diets are expensive and scarce, as a result, farmers resort to using a chicken starter ration containing 180 to 220 g/kg CP. At the beginning of the laying period higher protein intake will allow for a compensatory gain of the bird. For optimal performance, quails should be fed a diet containing approximately 12.0 MJ/kg of metabolisable energy and 10 g/kg calcium for the first six weeks (Randall & Bolla, 2008). Hyankova *et al.* (1997)

observed that quails consume 30 to 35 g per day but feed should always be available to them. For excellent performance, a game-bird diet can be fed but, it is very expensive and hard to find lastly, an adult quail requires (35 g) per day of feed, whereas an adult chicken requires (110 g), which means that farmers do not need to spend a lot of income buying large quantities of feeds.

2.5 Production systems

The choice of a production system is usually the farmer's preference. Farmers first consider their financial abilities, which primarily determine that production system they can manage. Time and space are also some of the factors that farmers have to consider before they choose a production system, for example the intensive production system needs daily or regular supervision, while the extensive production system requires too much land space for farming (Sonaiya, 2003). Just as in the production of broilers and laying hens, nutrition is one of the most important factors negatively impacting production costs of quail production, primarily due to the continuous fluctuation of prices of traditional dietary ingredients such as soybean meal inclusion, which stimulates increasing interest in possible alternative protein sources (Farahat *et al.*, 2013). Quail farming has evolved through three production systems, which are the traditional, small-scale semi commercial and large-scale commercial systems (Ravindran, 2013). These systems are based on a unique set of management styles and technologies. They differ markedly in terms of investment required, type of quails used, husbandry practices and inputs such as feed. The feed resources, feeding methods and feed requirements vary widely depending on the system used.

2.5.1 Extensive rearing system

The extensive rearing system is the most common type of quail production in developing countries. In this system, quails are often fed household or kitchen wastes, substances from the

surroundings (bugs, worms, veggies and seeds), crop residues, fodders and water plant life and commercial diets. The extensive poultry system is most beneficial for rural household free range production of meat, as most of the farmers need resilient chickens that can feed on home-made rations or fend for itself, stay alive and yield enough meat until it is slaughtered (Bosch, 2011). However, in areas with restricted natural resources, low rainfall, predation and diseases rearing may be difficult resulting in low productivity (Ravindran, 2013). Extensive rearing of quails plays a significant role in ensuring food security in disadvantaged areas of most developing nations, and is commonly practiced by communal farmers. It is also recommended for resource-poor farmers because quails are allowed to scavenge for their own feed without the provision of good shelter. This production system is inexpensive to carry out and is labour free so any community member can practice it.

Quails are omnivorous animals feeding on ants, bugs, kitchen leftovers and floor feed. This production system remains the favourite from the public as no antibiotics and/or remedies are used. The development of organic farming had been initiated due to public concerns relating to the utilization of antibiotic growth promoters, hormones and vaccination capsules. Most consumers prefer traditionally reared quails because their rearing management mimics that of organically farmed birds (Sanka & Mbagi, 2015). In this system, out of control breeding can escalate disease transmissions and inbreeding, leading to genetic defects and other abnormalities. Extensive farming addresses poverty and malnutrition in rural areas by ensuring that every household has a backyard stock of animals. Therefore, quails can be extensively reared as they require minimal input through cutting amount of feed that might be needed by chickens and this can contribute to poverty alleviation (Sonaiya, 2003; Bosch, 2012).

2.5.2 *High-input commercial systems*

This system is the most leading production units in both developed and developing countries, characterised by the incorporation of extremely sophisticated production units with high-performing animals (Karousa *et al.*, 2015). In intensive system, quails are reared indoors until slaughter or until the end of their production cycle, which means technical management is required. At all times, quails must receive a diet that meets their daily requirement and labour is essential to ensure feeding, cleaning and watering. The success of farming quails in a large-scale depends on the protein and energy content of the feeds, which is mostly derived from soybean and cereals. High productivity and efficiency rely on feeding nutritionally balanced feeds that are formulated to meet the quails nutritional requirements. Chowdhury *et al.* (2006) reported that good health and high productivity can increase with nutritionally balanced diets and proper management conditions. Economically developing countries invest on intensive quail production systems to produce meat and eggs for the growing human population.

2.5.3 *Semi-intensive farming*

The semi-intensive production system involves the rearing of quails confined to a large piece of land but having free access to natural pastures within the confinement. Semi-intensive production systems are characterised by small to medium flock sizes of 50 to 500 quails and birds are restricted overnight and are let out in the morning to scavenge (Ahlers *et al.*, 2009). Semi-intensive systems are commonly used by small-scale producers. This system require low expenditures but brings high returns, there is notable savings in feed costs accompanied by quality meat, which is lean and fat free compared to birds grown in intensive production systems ((Koster & Coetzee, 1996). However, growth and egg production are less when compared to quails reared intensively with better feed resources and well-balanced rations. Quails can only mate within the confinement and this system requires considerable amount of

labour. The shelter given is made from various materials, including wood and leaf material from local trees or shrubs. Losses may be experienced because of predation or theft, and failure to locate eggs that are laid in bushy areas, as a result, more labour is needed to manage flocks in the semi-intensive system compared to the intensive system (Ravindran, 2013).

2.6 *Moringa oleifera*

Moringa oleifera, commonly called horseradish tree or drumstick tree, is one of the most widely cultivated species of the monogenic family *Moringaceae*. Traditionally, *M. oleifera* is a pan-tropical tree that is known for its nutritional and medicinal properties in human nutrition. The *M. oleifera* tree was first introduced in Eastern Africa from India at the beginning of the 20th century (Adu-Dapaah *et al.*, 2017). It is widely distributed in different parts of the continent including Rwanda and Uganda in East Africa, Ghana in West Africa and South Africa. *Moringa* is a perennial softwood tree with timber of low quality due to its rapid growing nature (Makker & Becker, 1997; Sarwatt *et al.*, 2002). It is one of the world's most useful plants with nutritional and medicinal (nutraceutical properties) potential, water purifying and industrial uses (Fahey, 2005; Anwar *et al.*, 2007). *Moringa oleifera* leaf meal can be used as an alternative to some leguminous seeds as a source of high-quality protein (250 to 270 g/kg CP), oil and antioxidant compounds (phenols, vitamin C, vitamin E, β -carotene, zinc, selenium and flavonoids) to improve shelf-life and quality of meat products in the pre-slaughter or post-slaughter stages (Ferreira *et al.*, 2008; Valeria & Williams, 2011).



Plate 2.2. *Moringa oleifera* leaves and powder

Moringa oleifera leaf meal can cure many diseases and it has long been used in herbal medicine by Indians and Africans. The presence of phytochemicals in Moringa makes it a good medicinal agent (Bennett *et al.*, 2003). It can also be used as a way to treat water in rural areas where safe water resources are not available. It is a valuable source of livestock feed and its twigs are reported to be very palatable to ruminants and have appreciable crude protein levels (Sutherland *et al.*, 1990; Kimoro, 2002; Sarwatt *et al.*, 2002). *Moringa oleifera* is a valuable component of human and animal feed because of its amino acid profile, high level of vitamin A, E and its low level of ant nutritional compounds (Yang *et al.* 2006).

2.6.1 Nutritional composition of *Moringa oleifera*

The *Moringa oleifera* leaves contain important trace elements such as calcium, iron magnesium, zinc and copper (Kasolo *et al.*, 2010). Vitamins like β -carotene of vitamin A, vitamin B such as folic acid, pyridoxine and nicotinic acid, vitamin C, D and E are also present in *M. oleifera* (Mbikay, 2012). Phytochemicals such as tannins and alkaloids present along with anti-cancerous agents like glucosinolates, isothiocyanates, glycoside compounds and

glycerol 1-9-ocdecanoate (Berkovich *et al.*, 2013), thus qualifying the leaves as a potential animal feed supplement. Bennett *et al.* (2003), Ferreira *et al.* (2008) and Moyo *et al.* (2011) reported that MOLM contain 275 g/kg crude protein, 193 g/kg crude fibre, 223 g/kg crude fat and 713 g/kg 27.5% crude protein, 19.3% crude fibre, 2.23% crude fat and 7.13% ash. According to Mbikay (2012), the leaves have shown to control glucose metabolism in rats and humans, having a control over conditions such lipodema and hyperglycaemia, which renders cells susceptible to oxidation. Estrella *et al.* (2000) reported that MOLM increased breast milk production among young mothers of premature infants. Kakengi *et al.* (2003) reported that high pepsin and total soluble proteins in the leaves than other parts of the plant.

2.6.2 Bioactive compounds and medicinal properties of *Moringa oleifera*

Natural additives from plants are usually a desirable source for bioactive compounds that can be used as a replacement for AGPs, however, the only difficulty is their occurrence as complex mixtures instead of single compounds (Wallace *et al.*, 2010). Bioactive compounds are utilized for many purposes, but the conclusive objective is their advantageous interaction with the biochemistry of the body (Maiga *et al.*, 2005). Collectively, synergy between individual bioactive compounds in *Moringa oleifera* leaves may be a significant feature that affect broad components of physiology, nutrient absorption and immunity (Anwar *et al.*, 2007; Wallace *et al.*, 2010; Mbikay, 2012). Spirochin and anthonine are bioactive compounds found in the roots of *Moringa oleifera* and active against several bacteria. Moreover, *M. oleifera* leaves have bioactive phyto-constituent which lower cholesterol effects, β -sitosterol is capable of reducing cholesterol, suggesting that MOLM can reduce cholesterol levels in quail meat (Ghasi *et al.*, 2000).

High antioxidants present in *M. oleifera* leaves (Anwar *et al.*, 2007; Bamishaiye *et al.*, 2011; Mensah *et al.*, 2012) can be used to treat inflammatory conditions, cancer, hypertension and

heart diseases (Pari & Kumar, 2002; Posmontie, 2011). The extract of MOLM contains tannins, flavonoids and glycosides that has medicinal properties. These compounds have shown effective antioxidant, anti-carcinogenic and antimicrobial properties (Ayoola *et al.*, 2008; Davincelli *et al.*, 2015) due to their potential to inactivate lipid free radicals in phenolic compounds that are known to act as primary antioxidants and also prevent the decomposition of hydro peroxides into free radicals, due to their redox properties (Murillo & Fernandez, 2017). Furthermore, these properties play a pivotal role in neutralising free radicals, quenching of single or triple oxygen or decomposing peroxides (Pokorny, 2001; Zheng, 2001). Antimicrobial properties of MOLM have been validated after the inhibitory activity against several microorganisms (Ghasi *et al.*, 2000). In a recent study, aqueous extracts of MOLM were found to be inhibitory against many pathogenic bacteria such as *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa* (Saadabi & Zai, 2011).

Beside antibacterial activity of MOLM oils, it also has anti-fungal activity (Chuang *et al.*, 2007). *Moringa oleifera* leaf meal were found to be effective in preventing fungal growth of *Basidiobolus haptosporus* and *Basidiobolus ranarums* (Nwosu & Okafor, 1995). Pterygospermin can be used to counter the effects of a number of bacteria responsible for causing infections and also offers an anti-fungal effect for even more utility (Rao *et al.*, 1946). The seeds of *Moringa oleifera* plant include a natural antibiotics such as quercetin and β -carotene that can be used to treat or control infection. In addition, osmitrol activities of MOLM has been documented in its leaves, roots and seeds (Morton, 1991). Leaves of *Moringa oleifera* play part in controlling parasitic worms and their anthelmintic activity (Babar *et al.*, 2012; Hamad *et al.*, 2013).

2.6.3 Anti-nutritional factors in *Moringa oleifera*

Moringa oleifera is a popular plant species globally. This is owing to its usefulness as a food, medicinal, and aesthetic plant (Fuglie, 1999). However, like other plant species, the presence of anti-nutritional factors in this important species is capable of reducing the bioavailability of nutrients contained in the different parts. Makkar and Becker (1996) have reported on the presence of total phenols, tannins, saponins, phytate, lectins, cyanogenic glucoside and glucosinolate in different parts of *Moringa oleifera*. Similarly, Sallau *et al.* (2012) examined the effect of various processing methods on anti-nutrient content of *Moringa* leaves. Although anti-nutritional factors are generally understood to play negative roles by chelating the nutrients and forming a binding factor with the food values thereby making the nutrients non-bioavailable in the system, it is worth remarking that they also have beneficial effects at certain levels (Soetan & Oyewole, 2009).

2.6.3.1 Glucosinolates

Moringa oleifera plant is rich in compounds containing the simple sugar, rhamnose and a reasonably precise group of compounds known as glucosinolates and isothiocyanates (Fahey *et al.*, 2001; Bennett *et al.*, 2003). Glucosinolates are categorised as a volatile anti-nutritive compound that consists of antimicrobial, antifungal and antibacterial properties (Leeson & Summers, 2005). Four glucosinolates, two of which are found in huge concentrations in *M. oleifera* leaf tissues had been identified as 4-(α -l-rhamnopyranosyloxy)-benzylglucosinolate and three of its monoacetyl isomers (Bennett *et al.*, 2003). Numerous novel isothiocyanates are identified to hinder inducible nitric oxide synthase expression and nitric oxide synthesis in lipopolysaccharide stimulated RAW264.7 mouse macrophage cells (Cheenpracha *et al.*, 2011). This is further suggested to have anti-inflammatory and most cancers chemo-defensive ability (Park *et al.*, 2011). Absorption of glucosinolates may be harmful to animals' health and

production, the impact is more in non-ruminants as compared to ruminants, and greater in laying hens and turkeys compared to broilers, possibly due to their short life-span (Tripathi & Mishra, 2007).

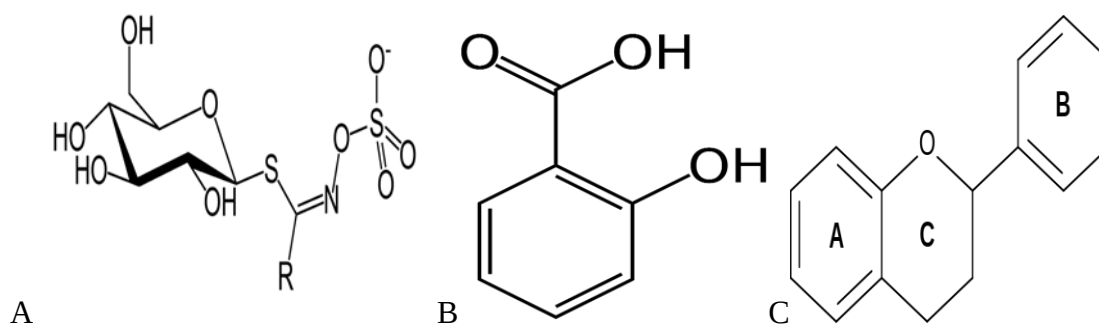


Figure 2.1. Structural motifs and backbones of major phytochemicals found in *M. oleifera* leaves. A = Glucosinolates; B = Phenols; C = Flavonols (Source: Mbikay, 2012).

2.6.3.2 Phenolics and flavonoids

Phenolic compounds are usually located in both palatable and non-palatable plants and they are said to have multiple organic effects, consisting of antioxidant activities (Kahkonen *et al.*, 1999). Two classes of phenolic acids can be distinguished, that is hydroxybenzoic acids (gallic acid, ellagic acids) and hydroxycinnamic acids (p-coumaric, caffeic, ferulic, chlorogenic and sinapic acids), which are more frequent in plants than the hydroxybenzoic acids (Kasetti *et al.*, 2012) and a lot of these had been removed in *M. oleifera* leaves (Mbikay, 2012). Newly opened *M. oleifera* leaves have the lowest phenolic content, which increases slowly with the maturity of leaves (Iqbal & Bhanger, 2006), hence they can be fed in a limited amount when leaves are matured.

Flavones are phenolic molecules containing one carboxyl group that yield flavonol on the addition of a three-hydroxyl organization, whereas flavonoids are hydroxylated phenolic substances that occur as a C6-C3 unit linked to an aromatic ring (Cowan, 1999). A sequence

of bioactive compounds such as flavonols and phenolic compounds have been identified in *M. oleifera* leaf extracts (Kahkonen *et al.*, 1999; Lemmen, 2007). Flavonols are a category of flavonoids that have the three-hydroxyflavone spine (IUPAC name: three-hydroxy-2-phenylchromen-4-1). Among the classes of phytochemicals determined inside the *M. oleifera* plant, flavonoids are reported to hold maximum of the antioxidant capacity (Mbikay, 2012). The predominant flavonols in *M. oleifera* leaves are quercetin and kaemferol, of their 3'-O-glycoside forms (Kahkonen *et al.*, 1999; Joshi & Meththa, 2010; Mbikay, 2012). Together with kaempferol rhanoside, kaempferol and quercetin-glucoside; other flavonol glycosides, namely, benzoic acid glycosides and benzaldehy de-glucoside, syringic acid, gallic acid and rutin had been isolated from the defatted aqueous methanolic extract of *M. oleifera* leaves (Manguro & Lemmen, 2007). According to Xiao *et al.* (2011) phenolics and flavonoids play a pivotal role with regards to their health benefits to animals as they have anti-inflammatory, antioxidant and anticancer properties.

2.6.3.3 Tannins and other anti-nutrients

Tannins are complex group of water soluble polyphenolic plant metabolites categorized as both condensed or hydrolysable and they have the potential to react with proteins, polysaccharides and different macromolecules (Wallace *et al.*, 2010). Condensed tannins (also known as proanthocyanidins) are structurally extra numerous and complex than hydrolysable tannins are derived from flavonoid monomers (Makkar & Becker, 1997). Condensed tannins are known to have a ability to combine with protein, carbohydrates, minerals and vitamins and they are known to be beneficial is livestock diets when they are at the concentration rate of 4 g/kg of the total diet intake (Mlambo *et al.*, 2015). Hydrolysable tannins include glucose as a significant core of polyhydric alcohol in addition to hydroxyl corporations, which are esterified

either partly or entirely with the aid of gallic acid to form gallotannins (Makkar & Becker, 1997).

Moringa oleifera leaves have 12 g/kg tannins in a DM basis and phytates at 21 g/kg (Makkar & Becker, 1997). According to Ferreira *et al.* (2008), phytate composition of 1-6% may decrease mineral bioavailability in monogastric animals, specifically Zn^{2+} and Ca^{2+} . Saponins are recognised to have beneficial cholesterol decreasing agents and toxic to intestinal cell toxicity and permeability (Bamishaiye *et al.*, 2011). Phytate are also implicated on the chelation of mineral ions and reduced digestibility of amino acids and carbohydrates, therefore impairing the provision of those compounds for poultry (Kornegay, 1999).

The ability of condensed tannins to bind proteins tempers with growth promotion an impact resulting from gallic acid and to a lesser quantity to the toxic compounds (pyroacetol and pyrogallol) (Leeson & Summers, 2005). Research have criticised tannins as a culprit that negatively affects feed intake, growth rate, feed efficiency, and protein digestibility in experimental animals (Chung *et al.*, 1998). However, the increasing research on tannins activities suggests that feed containing tannin components attract interests as alternative for antibiotic growth promoters in livestock and poultry feeding (Schiavone *et al.*, 2008). Furthermore, antimicrobial activity in tannins has been detected and can be associated with their ability to inactive microbial adhesions, enzymes and cell envelope transport proteins (Cowan, 1999). *Moringa oleifera* as a legume forage can also contain plant secondary compounds such as tannins and referred as ant nutritional components. These tannins have been re-valuated to better understand how this constituents influence animal health performance by improving feed intake, digestion and nutrients -use efficiency (Falowo *et al.*, 2018; Cwayita, 2012).

2.7 Climate change and livestock

Livestock production is likely to be adversely affected by climate change, global climate change is primarily caused by greenhouse gas emissions that results in warming of the atmosphere (IPCC, 2013). Climate change affects livestock production through competition for natural resources , quantity and quality of feeds, livestock diseases and heat stress (Garnett, 2009). Quality of feed crops and forage may be affected by increased temperature and dry conditions due to variations in concentration of water soluble carbohydrates and nitrogen. Temperature increases may increase lignin and cell wall components in plants (Polley *et al.*, 2013) which can reduce digestibility and degradation leading to a decrease in nutrient availability for livestock (Thornton *et al.*, 2009). Heat stress decreases forage intake, milk production, the efficiency of feed conversion and performance (Chase, 2012). There is a potential to reduce livestock sector greenhouse gas emissions through decreasing deforestation rates (Carvalho, 2004). Greenhouse gas can be also reduced by manure management, most methane emissions from manure management are related to storage and anaerobic treatment (Dickie *et al.*, 2014). Most mitigation practices involve shortening storage duration, improving timing and application of manure, used of anaerobic digesters and changing diets (ICF International, 2013).

2.8 Meat quality

The demand for high-quality protein source for human consumption is remarkably increasing, hence other alternatives such as quails rearing. The quality of poultry products determines their market value and its security for human nutrition (Ross-Tech, 2008). Quality of meat can be explained by its ability to provide all sorts of nutrients to the consumer. The tenderness and colour are some factors that determine the quality of the meat, although colour is usually to attract buyers. Producing rich-quality poultry products starts from the farm where the quails

are raised to the markets where they are processed and packaged for sales (Lin *et al.*, 2004). Most South Africans, particularly the old age, prefer organically produced poultry products for consumption (SAOFI, 2017), this can be attributed to the fact that no antibiotic growth promoters are used in this farming system. In addition, birds are extensively reared whereas in high-input intensive systems birds are modified (Petracci & Cavani, 2012). Although organic products are mostly preferred by many citizens, limited research have been conducted using natural additives to increase growth and production. The bioactive compounds of plants positively relate with biochemistry of the body and influences broader aspects of physiology, such as nutrient absorption and assimilation, and immunity (Anwar *et al.*, 2007; Wallace *et al.*, 2010; Falowo *et al.*, 2014). *Moringa oleifera* contain high levels of Sulphur-containing amino acids that can improve the detoxification process by donating methyl in various animal organs (Makkar & Becker, 1997). Detoxification method involves stimulating the liver to drive toxics from the body and improves circulation of blood. Substantial levels of α -tocopherol are encountered in MOLM at different growth stage during its maturity. The tocopherol content ranges from 5.7 mg/g for tender leaves to 27.8 mg/g mature leaves of dry mass (Sreelatha & Padma, 2009). The β -carotene, and vitamin A and C in these leaves supports their potential to include an antioxidant activity (Anwar *et al.*, 2007). Quercetin found in MOLM (Manguro & Lemmen, 2007), is a crucial glycoside compound that protects against cell degeneration by preventing lipid peroxidation (Joshi & Mehta, 2010). The presence of such antioxidants in *Moringa oleifera* have been reported to improve shelf life and the quality of carcass in the pre slaughter and post slaughter stages (Valeria & Williams, 2011).

2.8.1 Meat pH

Meat pH is primarily related to the biochemical state of the muscle at time of slaughter, following the development of rigor mortis (Wattanachant, 2008). Fletcher (1995) observed that

muscle pH and meat colour is highly correlated. Higher muscle pH is associated with darker meat, whereas lower muscle pH values are associated with lighter meat (Fletcher, 1999). High meat pH is often characterized as being dark, firm and dry meat and the lighter meat as being pale, soft and exudative (Wattanachant, 2008). The ultimate pH of meat is exceptionally structured upon the quantity of glycogen present inside the muscle, with the implication that pre-slaughter strain is related to muscle pH (Allen *et al.*, 1997). Consequently, the pre-slaughter feed withdrawal length has a capacity to have an effect on glycogen levels inside the muscle resulting in pH adjustments following rigor mortis (Lesiow & Kijowski, 2003).

2.8.2 Meat colour

According to Fletcher (1999) the colour of raw meat is critical for consumer selection, whereas the colour of cooked meat is critical for final evaluation. Meat and skin colour are influenced by various factors such as haeme pigments, feeding and genetics (Fletcher, 1999; Xiong *et al.*, 1999). Guan *et al.* (2013) stated that assessing meat quality by its colour is difficult due to genetic variations. Gordon and Charles (2002) found that slow-growing birds have a redder meat colour than fast growing birds due to age effect. The change in meat colour as chickens grow is always associated to structural changes in the muscle of the bird due to myoglobin as well as collagen and fats (Moran, 1999). Myoglobin in chickens normally increases with age, shifting the colour of the meat to dark and red tonalities (Fletcher, 2002). In contrast, Fanatico *et al.* (2005) reported that the meat of slow-growing birds has less red colouration than the fast-growing birds due to increased foraging of plant material. The physical appearance of meat is important to the consumers as this informs their choice of purchase. There are a number of factors that affect the physical appearance of meat such as genetics, ante- and post-mortem conditions, muscle chemistry and a number of factors related to meat processing and packaging

(Mancini & Hunt, 2005). Other major contributing factors to poultry meat colour are myoglobin content, the chemical state, and reactions of the myoglobin and meat pH.

The differences in colour, particularly the darker than normal colour ranges, may adversely have effects on final product colour (Fletcher *et al.*, 2000). Moreover, over the last decade the problem of the pale, soft and exudative (PSE) and its relevance to the poultry industry was given extensive attention by the industry and researchers (Barbut, 2009). Green plants such as MOLM are currently the main sources of β -carotene (Castañeda *et al.*, 2005). Carotenoids are compounds responsible for the pigmentation of skin, shanks and other non-feather tissues (Hamelin & Ultemueller, 2012). The rest of the body colour is influenced by the presence of the muscle pigments myoglobin and haemoglobin (Fletcher *et al.*, 2000). When myoglobin is exposed to oxygen the meat tend to become bright red and the meat may appear brown if myoglobin is exposed to small of oxygen or when the meat colour life is exhausted late in display when the iron in the pigment become oxidised (Harvey, 2008). Quails, however, cannot synthesize carotenoids, meaning that provision in the diet is crucial. Among the natural products, marigold meals and concentrates have been most widely accepted for commercial use in poultry feeds (Castañeda *et al.*, 2005). Traces of carotenoids, such as β -carotene (401 mg/kg DM) and xanhins (neoxanthin 219 mg/kg, violaxanthin 76.5 mg/kg, zeaxanthin 19.44 mg/kg) have been isolated in MOLM leaves. Xanthophylls, a class of carotenoids, are the most prominent source of pigmentation in poultry feeds (Hamelin & Ultemueller, 2012). The type of pigment used has implications in differences in metabolism or deposition, because even though synthetic pigments may show better absorption in the tissues; natural pigments have shown more efficiency in increasing skin yellowness (Castañeda *et al.*, 2005). Noteworthy, is the essentiality of carotenoids, not only for pigmentation, but also for immune-modulation as antioxidants—stream under immune stress periods, they drain drastically from the blood, resulting in reduced pigmentation throughout the whole body (Hamelin & Ultemueller, 2012).

2.8.3 Meat tenderness

Shear force is used to assess meat tenderness, and higher shear force means tougher meat quality (Cavitt *et al.*, 2004). Differences in muscle fibre size affect the shear force property of meat. Mahon (1999) reported that fast-growing chickens have longer fibre diameters than slow-growing lines. Longer fibre diameters are often associated with higher shear force and meat stiffening. Tang *et al.* (2009) stated that tenderness may be influenced by species, breed, age, sex, diet and individual skeletal muscle tissue of fowls. Kadim *et al.* (2003) highlighted that *Moringa oleifera* leaf meal diet can lead to high amount of intramuscular fats which can have a positive effects on tenderness.

2.8.4 Water holding capacity

The water holding capacity is term defines the water retention capacity of meat during the application of external forces such as cutting, grinding and pressing (Bowker & Zhuang, 2015). The extent of water loss depends on the ultimate pH reached, the higher the ultimate pH the less will be the loss of water, while the rate of decline of pH also determines the water retention of the meat. The higher the rate of pH drop, the greater the loss of water due to denaturation of proteins and also by the enhanced movement of the water into extracellular spaces. The colour, texture, juiciness and tenderness are affected by the amount of water (Fletcher, 1995).

2.8.5 Drip losses

From the point at which a bird is slaughtered during the meat production process, it is inevitable that water will be lost (drip loss) from the carcass. Water represents between 70% and 80% of the weight of raw poultry meat. The loss of water (drip loss) is a key concern for meat producers as this water content is said to contribute to the sensorial, organoleptic (juiciness, tenderness texture, smell and colour) and technological quality traits (Arango & Restrepo, 2003) of the

meat products. However, this impacts on consumer opinion, thus affecting demand and the saleable value (Prevolnik *et al.*, 2010; Maison *et al.*, 2016). According to Gil *et al.* (2008), drip loss is related to sensory qualities, such as hardness and juiciness. Muscles with a high drip loss have a high Warner Bratzler shear force. The results of a study by Mikulski *et al.* (2012) showed that a substitution of soybeans with a rapeseed meal at 120 g/kg increased the drip loss as compared to the control diet.

2.9 Summary

Quail farming is a new initiative in the South African poultry industry (Mnisi & Mlambo, 2018). However, if this industry can be sufficiently and sustainably developed, it can contribute enormously to animal protein supply, and this can guarantee food and nutrition security as well as economic growth of the country. Chickens have short generation intervals, high rate of reproduction and can efficiently transform nutrients into high quality protein (Akinfela *et al.*, 1999). Currently, poultry meat and eggs are by far the most consumed (FAO, 2000), accounting for more than 30% of all animal protein consumption (Permin & Pedersen, 2000). Quail meat is also known to have high in polyunsaturated fatty acids and essential trace minerals (Mnisi & Mlambo, 2018).

The use of antibiotics as growth promoters in the poultry industry has been banned in some jurisdictions due to concerns relating to harmful effects on human health. Consequently, herbs, spices, and various plant extracts, considered to be natural products that consumers would accept, have received increased attention as possible feed additives in place of antibiotic growth promoters (Catalagregori *et al.*, 2008). Natural additives from plants are usually a desirable source for bioactive compounds that can be used as a replacement for AGPs, however, the only difficulty is their occurrence as complex mixtures instead of single compounds (Wallace *et al.*, 2010). Several reports indicated that *Moringa oleifera* contain high crude protein, crude fibre,

crude fat and ash (Ferreira *et al.*, 2008; Moyo *et al.*, 2011) Kakengi *et al.* (2003) reported that high pepsin and total soluble proteins in the leaves than other parts of the plant. Different parts of *Moringa oleifera* also contain presence of total phenols, tannins, saponins, phytate, lectins, cyanogenic glucoside and glucosinolate (Makkar & Becker, 1996). Even though anti-nutritional factors are generally understood to play negative roles by chelating the nutrients and forming a binding factor with the food values thereby making the nutrients non-bioavailable in the system, it is worth remarking that they also have beneficial effects at certain levels (Soetan & Oyewole, 2009).

2.10 References

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3 CHAPTER THREE – GROWTH PERFORMANCE AND BLOOD INDICES OF JAPANESE QUAILS FED ON GRADED LEVELS OF *MORINGA OLEIFERA*

Abstract

The objective of the current study was to investigate the effect of dietary inclusion of *M. oleifera* leaf meal (MOLM) on growth performance and blood parameters of Japanese quails. A total of 224, 5-week old female Japanese quails were randomly but evenly distributed into 28 pens (experimental units) such that each pen held eight quails each. Four isonitrogenous and isoenergetic experimental diets formulated by diluting a commercial grower diet with *M. oleifera* leaf meal as follows were randomly allocated to the 28 pens and thus each diet was replicated seven times. The diets were formulated as follows: 1) control diet (a commercial grower diet) (CON), and 2) CON diluted with *M. oleifera* leaf meal at a rate of 25 g/kg, 50 g/kg and 75 g/kg (MOLM25, MOLM50 and MOLM75, respectively). Experimental diets and fresh water were offered ad libitum. Average weekly feed intake (AWFI) and average weekly weight gain (AWG) were used to calculate feed conversion efficiency (FCE). At the end of the 4 week feeding trial, all quails were slaughtered at a local abattoir. During slaughter, blood was collected from two random quails per experimental unit for determination of haematological and serum biochemical parameters. Repeated measures analysis indicated no significant week $\times$ diet interaction effect on AWFI and FCE. Dietary treatments had no significant effect on overall feed intake and overall FCE. However, there was a dietary effect on overall weight gain of quails. Japanese quails fed diet MOLM25 had the highest ($P < 0.05$) overall weight gain (30.72 g/bird) compared to those fed MOLM50 (18.60 g/bird), MOLM75 (22.76 g/bird) and CON (29.28 g/bird). Dietary treatments had no significant effect on all haematological parameters, except for haemoglobin. Japanese quails offered the MOLM25 diet had higher haemoglobin (14.85 g/dL) compared to those fed on MOLM75 (12.69 g/dL), MOLM50 (13.70

g/dL) and CON (14.32 g/dL). Experimental diets had no influence ($P > 0.05$) on all serum biochemical parameters of Japanese quails. There were significant dietary effects ($P < 0.05$) on the length of large intestine and weights of gizzards. Japanese quails offered MOLM75 (14.13 cm) had the longer large intestines compared to Japanese quails offered MOLM50 (11.24 cm), CON (12.73 cm) and MOLM25 (13.84 cm). On the other hand, diets had no effect ($P > 0.05$) on carcass characteristics and meat quality traits of Japanese quails. It was concluded that including MOLM at 25 g/kg improves weight gain and does not compromise physiological status of adult female Japanese quails.

Keywords: Blood indices, Feed additives, Growth, Japanese quails, *Moringa oleifera* leaf meal

3.1 Introduction

Quail farming plays a vital role in ensuring food and nutrition security because quails grow fast, attain sexual maturity earlier than other poultry birds, have short generation intervals, and are resistant to a number of avian diseases (Randall & Bolla, 2008; Mnisi *et al.*, 2017). However, due to high production costs as a result of using growth promoting antibiotics (AGPs) and other expensive feed ingredients such as maize and soy-based protein sources, quail farming is currently an unsustainable enterprise (Mnisi *et al.*, 2017; Mnisi & Mlambo, 2018). Historically, antibiotics have been used successfully as growth promoters in the poultry industry to increase growth rates through improving digestive health and nutrient utilization (Landy *et al.*, 2011). However, the continuous use of antibiotics resulted in build-up of antibiotic residues in meat products thus increasing health risks for consumers (Toghyni *et al.*, 2010). In addition, non-judicious use of antibiotics has given rise to antibiotic-resistant pathogens with the potential to affect human health directly.

Currently, there is no certified diet for quails in South Africa. Most quail farmers use expensive commercial feed formulated for exotic breeds, which may not meet or exceed the nutrient requirements of quails (Kingori *et al.*, 2010). As a result, it has become necessary to explore alternative natural additives such as *M. oleifera*, which can be a source of plant bioactive compounds such as carotenoids, vitamins, minerals, amino acids, alkaloids, and flavonoids. These bioactive compounds have the potential to boost nutrient utilization and reduce subclinical infection of the birds resulting in enhanced growth performance. *Moringa oleifera* has been reported to boost the immune system of broiler chickens (Jayavardhanan *et al.*, 1994; Fuglier, 1999; Olugbemi *et al.*, 2010). In evaluating the nutritive value of non-conventional feed resources such as *M. oleifera*, it is also important to assess the anatomical, physiological and health effects that such feed resources may have on the target animal. There are no studies that have investigated the utility of MOLM in quail diets. This study was, therefore, designed

to investigate the effect of graded levels of MOLM on growth performance, haematological and serum biochemical parameters in Japanese quails. It was hypothesised that the dilution of commercial grower diet with three graded levels of MOLM improves growth performance and alter blood metabolites of female Japanese quails.

3.2 Materials and methods

3.2.1 Study site and ingredient sources

This study was conducted at the North-West University Experimental Farm (Molelwane) with geographical coordinates of 25°40.459 S', 26°10.56'E, located in the semi-arid region of the North West province of South Africa. Ambient temperatures around the area ranges from 27°C to 37°C during summer, and from -3°C to 25°C during winter. The annual rainfall ranges from 300 to 600 mm. Fresh green *M. oleifera* leaf meal was supplied by Kingdom Business Investments (PTY) LTD (Gauteng, South Africa) with geographical coordinates of 25°51' 50.5" S 28°05' 19.9" E. Temperature of the area ranges from 18°C to 29°C during summer and between 5°C to 24°C. The average annual rainfall of the area is approximately 732 mm. and all the other ingredients were purchased from Opti Feeds (PTY) LTD (North West, South Africa).

3.2.2 Diet formulation

Four isocaloric and isonitrogenous diets were formulated by diluting a commercial grower chicken diet with graded levels of *M. oleifera* leaf meal (MOLM) using Format® nutritional software from Optifeeds (PTY) LTD (Lichtenburg, South Africa). The experimental diets were formulated as follows: 1. control diet (a commercial grower chicken diet) without *M. oleifera* leaf meal dilution (CON); 2. control diet diluted with *M. oleifera* leaf meal at a rate of 25 g/kg (MOLM25); 3. control diet diluted with *M. oleifera* leaf meal at a rate of 50 g/kg (MOLM50)

and 4. control diet diluted with *M. oleifera* leaf meal at a rate of 75 g/kg (MOLM75), as shown in Table 3.1.

Table 3.1. Ingredient and chemical composition (g/kg as fed basis, unless otherwise stated) of *Moringa oleifera* leaf meal-containing diets

	Diets ¹			
	CON	MOLM25	MOLM50	MOLM75
<i>Moringa oleifera</i> leaf meal	0	25	50	75
Yellow maize-fine	698.6	679.2	659.8	640.3
Prime gluten 60	18	18.0	18.0	18.0
Full fat soya meal	50.7	81.6	113.1	144.7
Soybean meal	196.7	160.2	123.1	86.1
Limestone powder	14.5	14.5	14.5	14.5
Mono calcium phosphate	7.2	7.2	7.2	7.2
Salt-fine	3.2	3.2	3.2	3.2
Sodium bicarbonate	1.7	1.7	1.7	1.7
Choline powder	0.8	0.8	0.8	0.8
Lysine	2.8	2.8	2.8	2.8
L- Threonine	0.4	0.4	0.4	0.4
Methionine	1.9	1.9	1.9	1.9
Grower-phytase	1.7	1.7	1.7	1.7
Premix	0.5	0.5	0.5	0.5
Olaquinox	0.4	0.4	0.4	0.4
<i>Chemical composition</i>				
Dry matter	901.6	903.1	904.6	906.0
ME (MJ/kg)	12.11	12.13	12.15	12.17
Crude protein	182.5	182.8	183.1	183.4
Crude fat	47.4	52.8	58.3	63.8
Crude fiber	25.8	26.8	27.7	28.6
Calcium	7.2	7.2	7.2	7.2
Phosphorus	4.5	4.5	4.4	4.4
Sodium	1.8	1.8	1.8	1.8
Potassium	7.3	7.3	7.3	7.2

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg

3.2.3 Chemical analyses

Subsamples of the formulated diets (CON, MOLM25, MOLM50, and MOLM75) were milled (Polymix PX-MFC 90 D) to pass through a 1 mm sieve for chemical analyses. For laboratory dry matter determination (AOAC, 2005; method no. 930.15), approximately 1 g of each leaf sample was placed into pre-weighed crucibles and placed in an oven set at 105°C for 12 hours. The loss in weight was measured as moisture content and DM was calculated as the difference between the initial sample and moisture weights. Organic matter content (AOAC, 2005: method no. 924.05) was determined by ashing the milled samples in a muffle furnace set at 600° C for 12 hours. The loss in weight was measured as organic matter (OM) content and the residue as ash. Total nitrogen content was determined by the standard macro-Kjeldahl method (AOAC, 2005: method no. 984.13) and was converted to crude protein by multiplying the percentage N content by a factor of 6.25. Amino acids determination was conducted by hydrolysing the samples with 6 N HCl (containing phenol) for 24 h at $110 \pm 2^\circ\text{C}$ in glass tubes sealed under vacuum as described by Ravindran *et al.* (2005). Crude fibre was determined using the ANKOM2000 Fibre analyser (ANKOM Technology, New York) with 0.2555 N crude fibre acid solution and then with 0.313 N crude fibre base solution. Crude fat and metabolisable energy (ME) contents were predicted using the near infrared reflectance spectroscopy (NIRs) Spectra Star XL (Unity Scientific, Australia).

Mineral content (calcium (Ca), phosphorus (P), sodium (Na) and potassium (K) of *M. oleifera* leaves was determined using the atomic ICP spectrophotometer (AAS-Buck 205) according to AOAC (2005) guidelines.

3.2.4 Sex differentiation and feeding trial

A total of 400, one-week old mixed-gender Japanese quails were purchased from Quails Breeders in Valhalla (Gauteng, South Africa). The quails were reared until five weeks of age

to allow gender differentiation using a commercial grower mash diet purchased from Opti Feeds (PTY) LTD (North West, South Africa). At five weeks of age, a total of 224 female Japanese quails were attained upon gender sexing. The females were randomly but evenly allocated to 28 pens (experimental units) each holding 8 quails. The size of each pen was 100 cm long × 60 cm wide × 30 cm high. The quails were allowed to adapt from to the pens and dietary treatments for a week before measurements commenced.

3.2.5 Feed intake and growth performance

Dietary treatments and clean water were provided *ad libitum* during feeding trial. Average weekly feed intake (AWFI) per bird per pen was measured from 6 to 10 weeks of age by subtracting the weight of the feed refusals from that of the feed offered and dividing the difference by the total number of quails in the pen. The initial live-weight of the quail per experimental unit was determined at the beginning of the experiment and thereafter measured weekly to determine average live weight by weighing all the quails. These live-weights were used to calculate the average weekly body weight gain (AWG) per bird as follows:

$$AWG (t_o, T) = \frac{W (T) - W (t_o)}{T - t_o}$$

Where t_o = initial time (days); T = final time (56 days); W (T) = final body weight/bird (g) and W (t_o) initial body weight/bird (g). Weekly feed conversion efficiency was calculated as:

$$FCE = \frac{\text{Weight gained (g)}}{\text{Feed consumed (g)}}$$

3.2.6 *Slaughtering and blood collection*

At 10 weeks of age, all Japanese quails were taken to Rooigrond poultry abattoir (North West province, South Africa). At the abattoir, birds were slaughtered humanely by electrical stunning and then all quails were live-hanged upside down onto a metal rack. Quails were then slaughtered by cutting the jugular vein with a sharp knife and they were left until bleeding ends. At slaughter, about 4 mL of blood was collected using syringes directly from the freshly severed jugular vein of two quails randomly selected from each pen. The blood was collected into two sets of sterilised tubes. Purple-top tubes containing ethylene diamante tetra acetic acid as an anti- coagulant were used for haematology, whereas red-top tubes without anticoagulant was used for serum biochemical analysis. For haematological analyses the samples were stored in a collar box with ice packs, whereas for serum biochemical analyses the samples were stored at a room temperature for a maximum of 45 min to clot and thereafter refrigerated at 4°C (Washington & van Hoosier, 2012).

3.2.7 *Blood analyses*

All analyses were conducted within 48 h after collection (Buetow *et al.*, 1999). Haematological parameters (erythrocytes, haemoglobin, hematocrits, leucocytes, lymphocytes, monocytes and neutrophils) were determined using an automated IDEXX LaserCyte Haematology Analyser (IDEXX Laboratories, Inc). Serum biochemical indices (albumin, alkaline phosphatase (ALP), alanine transaminase (ALT), amylase, serum cholesterol, blood glucose, lipase, total bilirubin and total protein) were analysed using automated IDEXX Vet Test Chemistry Analyser (IDEXX Laboratories, Inc).

3.3 Statistical analyses

Average weekly feed intake, average weekly weight gain and feed conversion efficiency data were analysed using repeated measures procedure of SAS (2010). The following statistical linear model was employed:

$$Y_{ijk} = \mu + D_i + W_j + (D \times W)_{ij} + E_{ijk}$$

Where Y_{ijk} = dependant variable, μ = population mean, D_i = effect of diets, W_j = effect of week, $(D \times W)_{ij}$ = effect of interaction between diets and week and E_{ijk} = random error associated with observation ijk , assumed to be normally and independently distributed.

Overall feed intake, overall weight gain, overall feed conversion efficiency, blood parameters data were analysed using the GLM procedure of SAS (2010) in completely randomized design (CRD). The linear statistical model was as follows:

$$Y_{ik} = \mu + D_i + E_{ik}$$

Where Y_{ij} = dependant variable, μ = population mean, D_i = effect of diets, and E_{ij} = random error associated with observation ij , assumed to be normally and independently distributed. For all statistical tests, significance was declared at $P \leq 0.05$. Least squares means (LSMEANS) were compared using the probability of difference option.

3.4 Results

3.4.1 Growth performance

Repeated measures analysis showed no significant week $\times$ diet interaction effect on weekly feed intake, weekly weight gain and weekly feed conversion efficiency. Table 3.2 shows that diets had significantly influenced overall weight gain but had no effect on overall feed intake and overall FCE. Japanese quails fed MOLM25 had higher ($P < 0.05$) overall weight gain (30.72 g/bird) whilst those on MOLM50 had the least weight gain (18.60 g/bird). Quails offered diet MOLM25 had similar ($P > 0.05$) overall weight gain with those offered diets CON and MOLM75. Similarly, overall weight gain in quails offered diet MOLM50 did not differ ($P > 0.05$) with those offered diets CON and MOLM75.

Table 3.2. Overall feed intake (g/bird), overall weight gain (g/bird) and overall feed conversion efficiency in Japanese quails fed graded levels of *Moringa oleifera* leaf meal

	Diets ¹				³ SEM	⁴ Significance
	CON	MOLM25	MOLM50	MOLM75		
Overall feed intake	847.3	871.7	870.6	830.6	32.8	NS
Overall weight gain	29.28 ^{ab}	30.72 ^b	18.60 ^a	22.76 ^{ab}	2.680	*
Overall FCE ²	0.139	0.141	0.088	0.109	0.014	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²Overall FCE: Overall feed conversion efficiency calculated over the entire duration of the study.

³SEM: Standard error of the mean.

⁴Significance: NS = not significant; * = $P < 0.05$.

^{a,b} In a row, dietary treatment means with common superscripts do not differ ($P > 0.05$).

3.4.2 Haematological parameters

Table 3.3 shows that there were no significant differences for all haematological parameters, except for haemoglobin. Erythrocytes ranged from 2.83 to $3.17 \times 10^{12}/\text{L}$, whereas haematocrit ranged from 0.437 to 0.506 L/L, while haemoglobin levels ranged from 12.69 to 14.85 g/dL. Leucocyte count ranged from 23.86 to $34.88 \times 10^9/\text{L}$, while lymphocyte counts ranged from 18.69 to $26.84 \times 10^9/\text{L}$ with monocyte counts ranging from 1.22 to $4.22 \times 10^9/\text{L}$. Neutrophil counts ranged from 4.31 to $11.05 \times 10^9/\text{L}$. Japanese quails offered the MOLM25 diet had higher haemoglobin (14.85 g/dL) compared to those fed diet MOLM75 (12.69 g/dL). Quails on MOLM25 had the same haemoglobin content ($P > 0.05$) as those on diets CON and MOLM50. However, quails on diet MOLM75 had similar haemoglobin content ($P > 0.05$) as those on diets CON and MOLM50.

Table 3.3. Effects of diets containing graded levels of *Moringa oleifera* leaf meal on haematological parameters of 10-week old Japanese quails

	Diets ¹				² SEM	³ Significance
	CON	MOLM25	MOLM50	MOLM75		
Erythrocytes ($\times 10^{12}/L$)	2.84	3.06	3.17	3.05	0.16	NS
Haematocrit (L/L)	0.44	0.51	0.50	0.50	0.02	NS
Haemoglobin (g/dL)	14.33 ^{ab}	14.85 ^b	13.71 ^{ab}	12.70 ^a	0.45	*
Leucocytes ($\times 10^9/L$)	23.87	34.88	32.47	24.68	4.31	NS
Lymphocytes ($\times 10^9/L$)	17.29	26.84	19.34	18.70	3.19	NS
Monocytes ($\times 10^9/L$)	3.03	1.22	4.22	3.34	0.75	NS
Neutrophils ($\times 10^9/L$)	3.31	5.61	11.05	4.31	3.23	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²SEM: Standard error of the mean.

³Significance: NS = not significant; * = $P < 0.05$.

^{a,b}In a row, dietary treatment means with common superscripts do not differ ($P > 0.05$).

3.4.3 Serum biochemical parameters

Table 3.4 shows that the experimental diet had no significant effect on all serum biochemical parameters of female Japanese quails. Alkaline phosphatase ranged from 298.6 to 820.9 IU/L whereas alanine transaminase ranged from 10.07 to 15.78 IU/L. Albumin ranged from 12.79 to 13.71 g/L while amylase ranged from 462.1 to 724.0 IU/L. Cholesterol ranged from 4.64 to 6.24 mmol/L whereas glucose ranged from 12.12 to 14.31 mmol/L. Lipase ranged from 40.7 to 63.78 IU/L, while total bilirubin ranged from 4.53 to 5.0 $\mu\text{mol/L}$. Total proteins ranged from 34.79 to 39.21 g/L.

Table 3.4. The effect of diets containing graded levels of *Moringa oleifera* leaf meal on serum biochemical parameters of 10-week old Japanese quails

	Diets ¹				² SEM	³ Sig
	CON	MOLM25	MOLM50	MOLM75		
Alkaline phosphatase (IU/L)	820.9	914.6	565.6	298.6	155.4	NS
Alanine transaminase (IU/L)	13.50	14.71	15.78	10.07	2.394	NS
Albumin (g/L)	13.71	12.79	13.21	13	0.608	NS
Amylase (IU/L)	724.0	497.4	563.2	462.1	102.6	NS
Cholesterol (mmol/L)	5.13	6.24	5.46	4.64	0.504	NS
Glucose (mmol/L)	14.31	13.57	12.12	13.44	0.979	NS
Lipase (IU/L)	57.5	63.78	56.21	40.71	6.97	NS
Total bilirubin (μ mol/L)	5.0	5.0	4.53	5.0	0.150	NS
Total protein (g/L)	39.21	34.79	36.78	37.71	1.61	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²SEM: Standard error of the mean.

³Significance: NS = not significant.

3.5 Discussion

Leaf meals are generally bitter in taste and tend to have higher levels of fibre, therefore, the inclusion of MOLM in the diets was expected to have resulted in reduced palatability, feed intake and growth performance of the quails. Indeed, Omekam (1994) observed that unpalatable feedstuffs have low intake in birds. Contrary to the expectations above, the inclusion of *M. oleifera* in diets of growing quails significantly ($P < 0.05$) enhanced weight gain with the maximum weight gain being observed in quails fed on MOLM25. This confirmed the observations made by Onu and Aniebo (2011), who reported that dietary supplementation with MOLM at 2.5% level significantly increased AWG of broilers. This could be related to the high nutritional content (Kangengi *et al.*, 2003; Sarwatt *et al.*, 2004) and antimicrobial properties of *M. oleifera* leaves (Fahey *et al.*, 2001). Similar findings were obtained by Nkukwana *et al.* (2014) who reported that supplementation of *M. oleifera* leaf meal up to 2.5% of feed did not impair nutrient utilization efficiency but enhanced the broilers's genetic potential for growth performance. In this study, adding moringa beyond 2.5% compromised growth performance of the quails, which can be due to higher levels of antinutritional factors (tannins, oxalate, saponins, phytates and fibre) that makes moringa leaves to have a bitter-taste rendering the diets-containing moringa unpalatable and poorly digested at higher inclusion levels (Gupta *et al.*, 1989; Tesfaye *et al.*, 2012). However, the performance of quails offered MOLM25 did not differ ($P > 0.05$) with those offered diets CON and MOLM75 in terms of overall weight gain, which was surprising given that MOLM was expected to improve weight gain of the quails due to the presence of growth-boosting bioactive compounds such as carotenoids, vitamins, minerals, amino acids, alkaloids, and flavonoids (Becker, 2003; Kakengi *et al.*, 2003; Sarwatt *et al.*, 2004). The lower weight gain in birds offered diets containing more than 25 g/kg MOLM could be attributed to higher crude fibre content, which is known to impair nutrient digestion and absorption by increasing the digesta viscosity (Aderemi, 2003; Onu & Otuma, 2008).

Haematology is usually used as indicators of the health status of the animal. According to Maxwell *et al.* (1990), blood parameters are important in assessing the quality and safety of feed ingredients for farm animals. Esonu *et al.* (2001) reported that haematological constituents reflect the physiological responsiveness of the animal to its internal and external environments, which include feed and feeding. Animashahun *et al.* (2006) affirmed that the comparison of blood chemistry profile with nutrient intake might indicate the need for adjustment of certain nutrients upward or downward for different population groups. There was no dietary influence on all hematological parameters of quails except hemoglobin, with all reported values falling within the normal ranges for adult female Japanese quails (Ali *et al.*, 2012; Mnisi *et al.*, 2017). This suggests that including moringa leaves in quail diets did not compromise the health status of the birds. The MOLM antimicrobial and antioxidant properties may be responsible for these findings, thus indicating that the experimental diets promoted the birds ability to wade-off subclinical infections (Ebenede *et al.*, 2012). Erythrocytes serve as a carrier for haemoglobin, reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration (Jonhston & Morris, 1996; Chineke *et al.*, 2006). A lower than normal haematocrit can indicate an insufficient supply of healthy red blood cells. Monocytes, lymphocytes, leucocyte and neutrophils are small white blood cells that plays a large role in defending the body against disease by attacking bacteria, viruses, and germs that invade the body (Chuang *et al.*, 2007). In the current study, there was significant dietary effect on haemoglobin. The higher haemoglobin counts were observed at inclusion level of MOLM25 leaf meal. The inclusion of MOLM25 leaf meal in Japanese quails' diets may have ensured sufficient iron intake, which promotes synthesis of haemoglobin (Ogbe & Affiku, 2012). Olugbemi *et al.* (2010) mentioned that red blood cells are responsible for the transportation of oxygen and carbon dioxide in the blood as well as manufacturing of haemoglobin, therefore, higher values indicate a greater potential for this function and a better state of health. *Moringa oleifera* leaf meal had no significant effect

on haematocrit values. Furthermore, Onu and Aniebo (2011) reported that haematocrit is an index of toxicity reduction in the blood usually and suggest the presence of a toxic factor, which has an adverse effect on blood formation. These results are in agreement with Hassan *et al.* (2016) who reported that MOLM supplementation affected haemoglobin concentration but not haematocrit counts.

Assessment of serum biochemical profile of quails provides useful information about their physical condition, making them useful tools in differentiating apparently healthy birds from abnormal and sick ones. Biochemical parameters can also be used to evaluate the nutritional status of birds. Although Japanese quails are fairly resistant to numerous avian diseases, serum biochemistry data can still be useful in diseases diagnosis and monitoring responses in quails, which often show no clinical signs (Fudge, 1997). Biochemical parameters are important in assessing the quality of feed ingredients in farm animals (Maxwell *et al.*, 1990), as they provide a good understanding of the nutritional value and toxicity of ingested feedstuffs in animals (Olafadehan, 2011). In the current study, there was no dietary influence on all serum biochemical parameters of quails, with all reported values falling within the normal ranges for adult female Japanese quails (Scholtz *et al.*, 2009; Ali *et al.*, 2012; Mnisi & Mlambo, 2018). Quails had the same blood protein concentration, this was expected since dietary diets were isonitrogenous and the partial changes of protein in the diet does not affect blood protein (Bovera *et al.*, 2007). Serum protein is a significant measure of the protein reserve in the animal body, suggesting that *M. oleifera* provided enough dietary protein (Akinola & Abiola, 1991; Esonu *et al.*, 2001). Albumin transports protein while glucose regulates osmotic pressure in birds, a higher concentration of albumin and total protein could be a sign of dehydration whereas lower concentrations are associated with malnutrition and infections (Esubonteng *et al.*, 2011). The levels of ALP were observed to be within the normal range for quails, suggesting that inclusion of MOLM did not introduce a large amount of toxic compounds that

could have induced abnormal liver function in a detoxifying effort. Bioactive compounds in MOLM are also known to protect against liver injury by restoring glutathione activities (Fakurazi *et al.*, 2008). These results are in agreement with Olugbemi *et al.* (2010), who reported that MOLM have no negative effect on the health status of broilers.

3.6 Conclusions

In this study, feed intake was at its peak when MOLM levels were between 25 and 50 g/kg suggesting that MOLM can be included at these levels to reduce feed costs and improve performance. Inclusion of MOLM in a commercial grower diet proved to have favourable effects on haemoglobin and had no negative effects on serum biochemical parameters suggesting that dietary use of the leaf meal for adult female Japanese quails does not compromise their pathophysiological status.

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4 CHAPTER FOUR- CARCASS CHARACTERISTICS AND MEAT QUALITY TRAITS OF JAPANESE QUAILS FED *MORINGA OLEIFERA*

Abstract

This study was conducted to assess the effect of *M. oleifera* leaf meal (MOLM) on carcass characteristics and meat quality of Japanese quails. A total of 224 female Japanese quails were randomly allocated to four isonitrogenous and isoenergetic experimental diets. The diets were formulated as follows: 1. commercial grower diet without MOLM (Control); 2. Control diet diluted with *M. oleifera* leaf meal at 25 g/kg (MOLM25); 3. control diet diluted with *M. oleifera* leaf meal at 50 g/kg (MOLM50) and 4. control diet diluted with *M. oleifera* leaf meal at 75 g/kg (MOLM75) and randomly allocated to 28 pens producing 7 replicates per dietary treatment as described in Chapter 3. At the end of the 4-week feeding trial described in Chapter 3, all quails were slaughtered at a local abattoir. After slaughter, the internal organs, carcass characteristics and meat quality data of quails were measured. Significant dietary effects ($P < 0.05$) were observed on length of large intestines and the weights of full and empty gizzards. Japanese quails offered MOLM75 had the longest large intestines (14.13 cm), which did not differ ($P > 0.05$) with those on CON (12.73 cm) and MOLM25 (13.84 cm). However, quails offered diet CON had similar ($P > 0.05$) size of large intestines to those offered diet MOLM50. The highest (5.45 g) gizzard weight was observed in Japanese quails fed MOLM75. The results on meat quality measurements also showed no significant dietary effect ($P > 0.05$) on meat pH, temperature, drip loss and shear force, water holding capacity and meat colour values of the breast muscle. It can be concluded that using *M. oleifera* leaf as feed supplements increased size of large intestines and gizzards but did not alter meat quality of Japanese quails.

Keywords: Bioactive compounds, *Moringa oleifera* leaf meal, carcass weight, internal organs, meat quality

4.1 Introduction

Poultry meat is among the cheapest sources of animal protein and is the most preferred and widely consumed meat worldwide (Le Bihan-Duval, 2004). The quality of the quail meat is defined by its suitability and acceptability to consumers and thus determines its market value. In addition, quality of meat is defined by its ability to provide sufficient nutrients to the consumer, as well as a desirable appearance and taste. Various factors, including the type of diet consumed by the animal, have been observed to affect meat quality parameters such as meat pH, water holding capacity, marbling, meat tenderness and meat colour. All these quality parameters are important in determining consumer preferences and meat purchasing decisions (Rimal, 2005). The meat industry as a whole is primarily driven by consumer preference, where bright red colour is seen as an indicator of freshness and overall wholesomeness (Risvik, 1994; Mancini & Hunt, 2005). The demand for high-quality protein for human consumption continues to rise while the cost of commercial feeds has become a major challenge in poultry production. A possible solution lies in the use of locally available, inexpensive, non-conventional, nutraceutical feedstuffs such as *M. oleifera*. *Moringa oleifera* leaves, due to their specific health and antioxidant properties, have received considerable attention as potential natural additives in livestock diets (Moyo *et al.*, 2011; Mbikay, 2012; Qwele *et al.*, 2013). Moreover, MOLM have high levels of sulphur- containing amino acids and are known to enhance the detoxification process by acting as methyl donors in various animal organs (Makkar & Becker, 1997). Bioactive compounds in *M. oleifera* leaves can regulate glucose homeostasis, stimulating its uptake and utilization by the liver, skeletal muscle, and adipose tissue through their modulation of the intestinal micro-flora (Mbikay, 2012). This process sustains the integrity of the immune cells that are beneficial in preserving animal health, and thus the quality of animal products (Frankie *et al.*, 2010; Wallace *et al.*, 2010). Pigments and antioxidants that are components of MOLM may enhance the colour and shelf-life of quail

meat. Research on the use of MOLM leaves to modify carcass traits and meat quality parameters of Japanese quails fed is scarce. Besides providing options for formulation of least cost diets, *M. oleifera* may desirably alter the meat quality traits for the benefit of consumers. This study was, therefore, designed to investigate the effect of graded levels of *Moringa oleifera* on carcass traits and meat quality parameters in Japanese quails. It was hypothesized that diluting commercial grower diet with MOLM will improve carcass traits and meat quality attributes of female Japanese quails.

4.2 Materials and methods

4.2.1 Study site and slaughter procedures

The study site was as described under Section 3.3.1 whilst the Section 3.3.2 and 3.3.4 explains the diet formulation and experimental design, which all apply in this chapter. The slaughter procedures are as described in the previous chapter (Section 3.3.6). After slaughtering the birds, they were put through a de-feathering machine. The birds were taken back to the North-West University Farm (Molelwane) laboratories for determination of internal organ size, carcass characteristics and meat quality attributes.

4.2.2 Carcass traits and internal organs

Carcasses from the different dietary treatments were identified by placing them in tagged plastic bags for each treatment. The feet, heads and intestines were removed by hand. The carcasses were then weighed at the abattoir to obtain the hot carcass weight (HCW). After 24 hours of chilling, they were weighed to obtain cold carcass weight (CCW). The dressing out percentage was calculated according to a standard procedure described by Uijttenboogaart and Gerrits (1982). The following organs were removed and weighed: gizzards, livers, hearts, and thighs, proventriculi and wings. The lengths of small and large intestines were measured and

recorded. Breast samples were collected 24 h post mortem for evaluation of the meat quality traits.

4.2.3 Meat pH and temperature

Meat pH and temperature were recorded immediately after slaughter (pH_i and Temp_i) and 24 hours post slaughter (pH₂₄ and Temp₂₄) on the breast muscle (central area of the breast) using a Corning Model 4 pH-temperature meter (Corning Glass Works, Medfield, MA) equipped with an Ingold spear-type electrode (Ingold Messtechnik AG, Udorf, Switzerland) (Standford *et al.*, 2003). After every 20 measurements, the pH meter was calibrated with standard solutions (Ingold Messtechnik AG, Udorf, Switzerland).

4.2.4 Meat colour

Colour of the meat (L^* = Lightness, a^* = Redness and b^* = Yellowness) was determined using a Minolta colour-guide (BYK- Gardener GmbH, Geretsried, Germany), with a 20 mm diameter measurement area and illuminant D65-day light, 10° observation angle. The colour meter was calibrated before measurements using the green standard. Colour recording was done on the surface of the thigh muscle allowed to bloom for 1 h on a polystyrene tray at 4°C. Hue angle, $\tan(\theta) = \frac{a^*}{b^*}$, and chroma was calculated as $\sqrt{a^{*2} + b^{*2}}$ as guided by Priolo *et al.* (2002).

4.2.5 Meat cooking losses

After weighing, breast samples were placed in an oven set at 140°C for 20 minutes for determination of cooking losses (Gopinger *et al.*, 2014). The following formula was used:

$$\text{Cooking losses(\%)} = \frac{\text{initial weight} - \text{final weight}}{\text{initial weight}} \times 100$$

4.2.6 Meat tenderness

After cooking, breast samples were sheared perpendicular to the fibre direction using a Warner-Bratzler shear device mounted on a Universal Instron apparatus (cross head speed= 200 mm/minute, one shear in the centre of each core). The reported value represented the average shear force measurements of each sample in Newtons.

4.2.7 Water holding capacity

The WHC of the meat measurements were done in duplicate samples on the surface of a freshly cut slice of the pectorals major muscle (PMM) (8-16 g) and were determined as the amount of water expressed from fresh meat held under pressure (60 kg pressure) using the Filter-Paper Press method developed by Grau and Hamm (1957). Water holding capacity was calculated using the equation:

$$\text{WHC (\%)} = \frac{\text{initial weight} - \text{weight after pressing}}{\text{initial weight}} \times 100$$

4.2.8 Drip loss

Drip loss measurement was determined using a method adapted from Zhang *et al.* (2009). Briefly breast meat samples were sliced with a knife into blocks weighing between 2-3 grams. The sample weights were recorded as initial weight (W_1). The sample were hooked and suspended using wire steel in a plastic and sealed properly so that the samples did not touch the sides of the bottle. The bottles were then stored in cold room 4°C for 72 hours, the samples were taken out gently wiped to remove any liquid on the surface of the meat and the sample were reweighed (W_2). The difference in weight of each sample before and after drip was conveyed as percentage drip loss and calculated as follows:

$$\text{Drip loss (\%)} = \left[\frac{(W1 - W2)}{W1} \right] \times 100$$

4.3 Statistical analyses

Data on carcass characteristics and meat quality traits were analysed using general linear models (GLM) procedures of SAS (2010) in completely randomized design (CRD). The linear model was as follows:

$$Y_{ij} = \mu + D_i + E_{ij}$$

Where, Y_{ij} = observation of the dependent variable (carcass characteristics and meat quality traits), μ = population mean constant common to all observations, D_i = effect of diets, and E_{ij} = random error associated with observation ij , assumed to be normally and independently distribute. For all statistical tests, significance was declared at $P \leq 0.05$. Least squares means (LSMEANS) were compared using the probability of difference (PDIFF) option in the LSMEANS statement of SAS.

4.4 Results

4.4.1 Visceral organs

Table 4.1 shows that the size of full gizzard, empty gizzard and large intestines was influenced ($P < 0.05$) by the experimental diets. Japanese quails on MOLM75 had heavier gizzards (3.87 g), than quails offered MOLM25 (3.25 g). Quails fed MOLM75 did not differ ($P > 0.05$) with those on CON (3.37 g) and MOLM50 (3.34 g). However, the weight of gizzards in quails fed CON was similar ($P > 0.05$) to those fed MOLM25 and MOLM50. Japanese quails offered MOLM75 had the longest large intestines (14.13 cm) than quails offered MOLM50 (11.24 g) Japanese quails fed MOLM75 did not differ ($P > 0.05$) with those on CON (12.73 cm) and MOLM25 (13.84 cm). However, quails offered diet CON had similar ($P > 0.05$) size of large intestines to those offered diet MOLM50. Diets had no effect on the size of the hearts, livers, proventriculi and small intestines. The hearts ranged from 1.54 to 1.96 g, while the liver ranged from 3.82 to 4.57 g. The small intestines ranged from 43.89 to 47.17 cm, whereas proventriculi ranged from 0.786 to 0.856 g.

Table 4.1. Size of internal organs of 10-week old Japanese quails fed graded levels of *Moringa oleifera* leaf meal containing diets

	Diets ¹				² SEM	³ Significance
	CON	MOLM25	MOLM50	MOLM75		
Gizzards (g)	3.37 ^{ab}	3.25 ^a	3.34 ^{ab}	3.87 ^b	0.131	*
Heart (g)	1.89	1.54	1.59	1.96	0.141	NS
Large intestine (cm)	12.73 ^{ab}	13.84 ^b	11.24 ^a	14.13 ^b	0.622	*
Liver (g)	4.56	4.47	3.82	4.57	0.285	NS
Proventriculi (g)	0.855	0.786	0.801	0.856	0.041	NS
Small intestine (cm)	45.37	45.15	43.89	47.17	1.36	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²SEM: Standard error of the mean.

³Significance: NS = not significant; * = $P < 0.05$; ** = $P < 0.01$.

^{a,b} In a row, dietary treatment means with common superscripts do not differ ($P > 0.05$).

4.4.2 Carcass characteristics

Table 4.2 shows that there were no dietary influence ($P > 0.05$) on all carcass characteristics of Japanese quails. Slaughter weight ranged from 205.6 to 226.1 g, while HCW ranged from 127.8 to 142.4 g. The CCW weights ranged from 126.1 to 139.7 g, whereas the dressing out percentage ranged from 58.71 to 65.93%. The length of the thighs ranged from 4.89 to 5.03 cm, while the weights of the thighs ranged from 5.56 to 6.37 g. The length of the wings ranged from 10.08 to 10.26 cm while the weights of the wings ranged from 5.41 to 6.35 g.

Table 4.2. The effect of graded levels of *Moringa oleifera* leaf meal on carcass characteristics of 10-week old Japanese quails

Trait ²	Diets ¹				³ SEM	⁴ Significance
	CON	MOLM25	MOLM50	MOLM75		
SW (g)	226.1	223.9	205.6	212.0	6.08	NS
HCW (g)	142.4	130.6	127.8	139.4	5.03	NS
CCW (g)	139.7	127.7	126.1	134.0	4.62	NS
Dressing out (%)	62.70	58.71	62.22	65.93	2.02	NS
Thighs (cm)	5.01	5.03	4.89	5.03	0.114	NS
Thighs (g)	6.37	5.91	5.56	5.90	0.213	NS
Wings (cm)	10.26	10.08	10.20	10.20	0.172	NS
Wings (g)	6.35	5.79	5.41	5.73	0.323	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²Parameters: SW = slaughter weights; HCW = warm carcass weights; CCW = cold carcass weights.

³SEM: Standard error of the mean. ⁴Significance: NS = not significant.

4.4.3 Meat quality

Table 4.3 shows that for meat quality parameters measured immediately after slaughter, experimental diets had no influence ($P > 0.05$). Meat pH_i ranged from 6.96 to 7.23, meat temperature ranged from 23.0 to 29.08 °C, meat lightness (L^*) ranged from 48.17 to 49.77, meat redness (a^*) ranged from 1.25 to 1.57, meat yellowness (b^*) ranged from 10.81 to 11.53, meat Chroma ranged from 4.82 to 5.08 and hue angle ranged from 1.42 to 1.48 of female Japanese quails.

Table 4.3. The effect of graded levels of *Moringa oleifera* leaf meal on meat quality parameters of 10-week old Japanese quails immediately after slaughter

	¹ Diets				SEM ²	Significance ³
	CON	MOLM25	MOLM50	MOLM75		
Meat pH _i	6.96	7.09	7.23	7.01	0.096	NS
Temp _i (°C)	24.40	29.08	28.06	23.0	2.29	NS
<i>L</i> *	48.51	49.77	48.51	48.17	0.829	NS
<i>a</i> *	1.25	1.02	1.57	1.43	0.197	NS
<i>b</i> *	10.81	11.18	11.08	11.83	0.456	NS
Chroma	4.82	4.85	4.99	5.08	0.114	NS
Hue angle	1.46	1.48	1.42	1.45	0.015	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²Parameters: *L** = lightness ; *a** = redness; *b** = yellowness; pH_i = initial pH; temp_i = initial temperature.

³SEM: Standard error of the mean; ³Significance: NS = not significant.

Table 4.4 shows that for meat quality parameters measured 24 h post slaughter, experimental diets had no significant effect. Meat pH_u ranged from 6.65 to 7.62, meat temperature ranged from 13.19 to 13.64°C meat lightness (*L**) ranged from 42.41 to 48.71, meat redness (*a**) ranged from 4.19 to 6.44, meat yellowness (*b**) ranged from 8.74 to 10.58, meat chroma ranged from 5.11 to 5.58 and hue angle ranged from 1.04 to 1.14 of female Japanese quails.

Table 4.4. The effect of graded levels of *Moringa oleifera* leaf meal on meat quality parameters of 10-week old Japanese quails after 24 h slaughter

Trait ²	¹ Diets				SEM ²	Significance ³
	CON	MOLM25	MOLM50	MOLM75		
Meat pH _u	6.65	7.62	6.70	6.67	0.524	NS
Meat temp _u (°C)	13.19	13.39	13.64	13.21	0.367	NS
<i>L</i> *	48.71	42.41	48.50	48.30	3.70	NS
<i>a</i> *	4.19	4.78	6.44	4.96	1.05	NS
<i>b</i> *	9.57	9.05	10.58	8.74	1.19	NS
Chroma	5.19	5.11	5.58	5.22	0.293	NS
Hue angle	1.12	1.14	1.04	1.05	0.052	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²Trait: *L** = lightness ; *a** = redness; *b** = yellowness ; pH_u = ultimate pH; temp_u = ultimate temperature.

³SEM: Standard error of the mean; ³Significance: NS = not significant.

There was no significant dietary effect on cooking loss (43.19 - 44.80%), drip loss (18.45 - 20.41%), shear force (5.77 - 5.95 N) and water holding capacity (5.19 - 7.59%) values of Japanese quails (Table 4.5).

Table 4.5. The effect of graded inclusion levels of *Moringa oleifera* leaf meal on meat quality parameters of 10-week old Japanese quails

	¹ Diets				³ SEM	⁴ Significance
	CON	MOLM25	MOLM50	MOLM75		
Cooking loss (%)	44.80	43.37	43.19	43.95	1.407	NS
Drip loss (%)	19.26	19.30	20.41	18.45	1.427	NS
Shear force (N)	5.92	5.77	5.84	5.95	0.583	NS
² WHC (%)	7.14	7.59	6.46	5.19	1.25	NS

¹Diets: CON = control diet (a commercial grower diet without MOLM); MOLM25 = control diet diluted with MOLM at a rate of 25 g/kg; MOLM50 = control diet diluted with MOLM at a rate of 50 g/kg; MOLM75 = control diet diluted with MOLM at a rate of 75 g/kg.

²WHC: Water holding capacity.

³SEM: Standard error of the mean.

⁴Significance: NS = not significant.

4.5 Discussion

Birds respond quickly to changes in dietary fibre content as seen by changes in the intestinal length and weight of internal organs (Mateos *et al.*, 2012). These adaptation changes are coping strategies that might result in improvements in nutrient digestibility, gastrointestinal tract health, animal welfare and, eventually, growth performance (Kalmendal *et al.*, 2011). In the current study, quails offered MOLM75 had longer large intestines compared to those offered MOLM25, which was expected because diet MOLM75 has more fiber thus expected to have longer large intestines than quails on MOLM50. This is in agreement with Borin *et al.* (2005) as well as Rubio and Brenes (2010), who indicated that high proportions of undigested substances in the digesta increased the lengths of the intestinal sections. Longer intestines are assumed to digest feed efficiently and provide greater surface area for nutrient absorption. Results on size of gizzards and large intestines in birds offered MOLM25 and MOLM50 contradict findings in previous studies where no differences on viscera macromorphometry were observed (Shaba *et al.*, 2016). Musa *et al.* (2006) indicated that benefits of a larger gizzard (MOLM75) include improved muscular grinding of feed particles leading to higher digestibility of nutrients. In addition, authors also reported that consumption of feed with high fibre content result in the enlargement of the gizzard. This is because fibre particles are in general difficult to digest than other dietary components so they tend to gather in the gizzard (Hetland *et al.*, 2005) thus stimulating muscular development and functioning of this organ. Diets had no significant effects on the size of liver, heart, breast, proventriculi weights and small intestines. These findings are in agreement with Hassan (2016), who reported no significant differences among dietary treatments in liver, heart, heart, proventriculi and small intestine of broilers when fed MOLM supplements. The liver contains enzymes such as ALT and ALP, these enzymes are released into the bloodstream when damaged (Sherwin, 2003). Elevation of these serum enzymes occur with conditions of altered hepatocellular membrane

permeability either due to circulatory hypoxia, exposure to toxins and toxaemia, inflammation, metabolic disorders or proliferation of the hepatocyte. In the stunting syndrome of Japanese quails, increased activities of ALP and ALT were also associated with liver and intestinal damage (Rani *et al.*, 2011). Hence, the normal levels of these enzymes even in quails receiving MOLM-containing diets reflects the normal liver and intestinal functions of quails suggesting that the leaf meal is a safe dietary ingredient for these birds.

Feeding MOLM-containing diets had no effect on carcass traits. There was no significant ($P > 0.05$) difference in carcass dressing percentage across all the dietary treatments (Table 4.2). The weights of thighs and wings of quails were also found to be similar across experimental diets. The present results confirmed the findings of Safa and Tazi (2012), who indicated that none of the parameters measured for carcass characteristics in birds fed with diets containing *M. oleifera* leaf meal were significantly affected. In addition, Ayewiede *et al.* (2011) and Odetola *et al.* (2012) also reported that MOLM did not significantly influence the carcass characteristics (SW, HCW, CCW, dressing out, thighs and wings) in poultry. This finding is also consistent with the findings of Cui *et al.* (2018), who reported no significant responses on carcass traits in broilers fed MOLM-containing diets. According to Brickett *et al.* (2007); Havenstein *et al.* (2003) and Young *et al.* (2001), carcass yield is affected by genetic, feed, slaughtering conditions, body weight and gender of birds. In addition, low body weight is associated with low carcass yield (De Marchi *et al.*, 2005; Fanatico *et al.*, 2005; Moujahed & Haddad, 2013).

Meat pH value is one of the most important factors that influence meat quality parameters such as meat cooking loss, tenderness, drip loss and WHC (Allen *et al.*, 1998). Husak *et al.* (2008) reported that higher pH meat is more effective for retaining a desirable colour and moisture absorption properties. The ultimate pH indicates the extent of pH decline after slaughter. The range is determined by how much glycogen is in breast muscle prior to slaughter and how

rapidly the remaining glycogen is converted to lactic acid after slaughter (Dyubele *et al.*, 2010). In this study, diets had no significant effect on meat pH, meat colour (L*, a*, b*, chroma, and hue angle), shear force, cooking loss, drip loss and water holding capacity measured 24h post mortem. The lack of effects indicates the potential of moringa to promote normal oxidative stability for meat quality attributes during storage, which was consistent with the findings of Qwele *et al.* (2013), who reported no significant responses on carcass traits in broilers fed MOLM-containing diets. The pH_u of the meat after slaughter across all dietary treatments was however, higher than the recommended pH for good quality meat, which is usually from 5.4 to 5.7 (Jaturasitha *et al.*, 2008). The higher pH values are associated with dark, firm and dry (DFD) (Swatland, 2008). Bai *et al.* (2013) reported that due to post-mortem glycolytic activity that results in lactic acid reduction in muscle, meat pH tends to increase significantly. The colour of meat is important to consumers when buying meat products and is an important factor affecting consumer acceptance of meat and meat products (Muchenje *et al.*, 2009, Xazela *et al.*, 2012). In the current study diet did not have any effects on lightness of the meat as observed in other studies (Mnisi *et al.*, 2017). Diet did not affect the colour of the meat, which was not expected given the carotenoids and other pigments present in MOLM but it could also be that at 2.5, 5.0 and 7.5% inclusion rates, the absolute quantities of the pigments were not sufficient to induce colour changes. In this study, the L* values of the breast muscle were within the normal range suggesting that the meat produced cannot be considered to be excessively pale (Jaturasitha *et al.*, 2008; Laudadio & Tufarelli, 2010; Motsepe, 2016). The redness (a*) values were in the normal range of 4.19 - 6.44 obtained for Japanese quails as reported by Genchev (2008). The yellowness (b*) values in the present study also fell within the normal range as reported by Genchev (2008). Although diet had no effect on both drip loss and cooking loss, it has been observed that generally, higher drip loss may induce a reduction in water-holding capacity and tenderness of meat (Wang *et al.*, 2012). The lowest level of water loss was

observed at higher inclusion levels of MOLM, this may be an indication of better water retention compared to control diet. The shear force is an objective indicator of the toughness of the meat (Lonergan *et al.*, 2003) with a lower shear force value indicating meat that is tenderer (Lonergan *et al.*, 2003; Li *et al.*, 2016). However, in this study there was no dietary influence on the shear force values although the shear force values tended to increase with MOLM inclusion suggesting that increasing dietary MOLM inclusion may have rendered the meat tougher.

4.6 Conclusions

Inclusion of *M. oleifera* leaf meal increases the size of gizzards and large intestines in Japanese quails. However, the inclusion of *M. oleifera* did not affect carcass characteristics and meat quality measures of Japanese quails.

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5 CHAPTER FIVE - GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

5.1 General discussion

The growth of human and livestock populations has increased the competition for food and feed between animals and man (Odunsi, 2003), suggesting that researchers must find more alternative feedstuffs to decrease the competition between animals and man. Poultry production is probably the fastest means of meeting the rising demand for animal protein supply on the African continent. This is due to their short generation interval, high reproduction rate and nutrient transformation efficiency into high quality protein (Akinfela *et al.*, 1999). However, considering how fast the human population is growing, there is a need to redirect attention to the production of other fast-growing avian species such as Japanese quails on a commercial scale. Japanese quails (*Coturnix coturnix japonica*) have emerged as new entrants into the poultry industry in many developing countries and around the world (Khosravi *et al.*, 2016). Quails have been shown several attributes such as disease resistance, tolerance to harsh environmental conditions, and early sexual maturity at 6-weeks of age and fast growth rates (Ayasan *et al.*, 2011; Khosravi *et al.*, 2016; Cong *et al.*, 2017). These qualities make the quails an appropriate bird species for intensive food production. This has resulted in intense efforts to improve their production in order to ensure sufficient nutrition for the fast-growing human population. Therefore, utilization of alternative feed resources such as *M. oleifera* that cannot be utilised directly as food for humans must be fully explored. To this end, a feeding trial was conducted over a period of 4 weeks to determine the effects of dietary inclusion of *M. oleifera* leaf meal on growth performance, haematology, serum biochemistry, carcass characteristics and meat quality parameters of Japanese quails. The MOLM was chemically analysed and used to dilute a commercial poultry grower diet at 0 (CON), 25 (MOLM25), 50 (MOLM50) and 75

(MOLM75) g/kg DM, producing four isonitrogenous and isoenergetic diets, which were fed to 224 Japanese quails. At 10 weeks of age blood samples were taken during slaughter from two quails per experimental unit and used for haematological and biochemical analysis.

In chapter 3 of this thesis, a study was designed to determine the growth performance and haemo-biochemical parameters of Japanese quails fed graded levels of MOLM. It was observed that there were significant dietary differences in terms of growth performance. Dietary inclusion of MOLM above 25 g/kg was found to have reduced voluntary feed intake and thus compromising the performance of female Japanese quails. This could also be due to the negative effect of the antinutritional factors present in *M. oleifera* leaf meal on the quails. Furthermore, leaf meals are generally bitter in taste, therefore, the inclusion of MOLM in the diets was expected to result in reduced palatability and feed intake. In this study, the inclusion of *M. oleifera* leaf meal at 2.5% promoted weight gain of female Japanese quails, suggesting that MOLM should be used in adult female quail diets and at this level, serum biochemical parameters are indicators of pathophysiological state of level, thus their determination in Japanese quails fed MOLM-containing diets was inevitable to safeguard the wellbeing of the birds. Indeed, haemoglobin content was significantly influenced by dietary treatments with the highest level being observed in MOLM25 quails. This suggests that inclusion of *M. oleifera* leaf meal in Japanese quail diets may have increased iron intake, which stimulates the synthesis of haemoglobin and increases the production of red blood cells. Since most haematological and serum biochemical parameters fall within the expected normal ranges for quails, it means that inclusion of MOLM in quail diets did not negatively affect the health and nutritional status of the birds.

In chapter 4, dietary treatments were also found to have no significant effect on all carcass characteristics, thus the proposed alternative hypothesis that feeding MOLM-containing diets to quails would alter these parameters was rejected. However, only the size of large intestines

and gizzards were significantly influenced by dietary treatments, which was thought to be the result of relatively higher fibre levels in MOLM-containing diets. However, the proventriculi, liver, heart and small intestine and sizes were not markedly affected by dietary treatments. The results on meat quality measurements also showed a lack of significant effect of diet. These results also led to the rejection of the proposed alternative hypothesis, which stated that MOLM inclusion in quail diets would alter the pH, temperature, hue angle, cooking loss, drip loss, tenderness and WHC of meat when measured 24 h post mortem. The lack of effects indicates the potential of MOLM to promote normal oxidative stability for meat quality attributes during storage.

5.2 Conclusions

The results from the feeding trial showed that *M. oleifera* inclusion increased haemoglobin levels, improved growth performance, and increased the sizes of gizzards and large intestines. It was hypothesized that the inclusion of *M. oleifera* leaf meal alters physiological and meat quality responses of Japanese quails. This suggests that quail diets containing MOLM have the potential to improve productivity of Japanese quails in a cost-effective and safe way. Moreover, *M. oleifera* utilization in poultry diets can be introduced for the small scale farmers as a natural and healthy feedstuff alternative. From the current results, it is recommended that *M. oleifera* leaf meal be included at 2.5% level for the rearing of quail birds.

5.3 Future research

Research on the growth performance of Japanese quails fed on graded levels of *M. oleifera* meal with multi-enzyme supplementation should be conducted in order to assess the performance of quails when compared to current results.

One major limitation on quails digestion is the fact that they do not produce enzymes that can break down fibre, suggesting that less fibrous diets should be provided to ensure optimal performance. Again, due to change of environment from the collection site to the experimental area, mortality might occur. Mortality might also be experienced during diet adaptability period.

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5.4 References

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