The evaluation of lupins (*Lupinus angustifolius*) as alternative protein source to soybean oilcake meal in ostrich (*Struthio camelus* var. *domesticus*) diets

By

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Declaration

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Abstract

In an intensive ostrich production unit, one of the highest cost components is nutrition, contributing *ca*. 75% to the total input costs. Energy and protein are the two most important and abundant nutrients found in a balanced diet, with the protein component being the most expensive per unit weight. This study was conducted to evaluate the inclusion of lupins (*Lupinus angustifolius*) as an alternative protein source to soybean oilcake meal in ostrich (*Struthio camelus* var. *domesticus*) diets.

In the first study (chapter 3) production performance and slaughter traits were studied at different dietary lupin inclusion levels that replaced soybean oilcake meal. There were five dietary treatments with three replications of between 15 – 17 chicks each. The chicks all received the same pre-starter diet. Five iso-nutritional diets were formulated for each feeding phase (starter, grower and finisher) using Mixit2+ software according to specifications set out for each of the different feeding phases Within each feeding phase these diets contained either soybean oilcake meal (control diet; 0LD) or sweet lupins (alternative protein source; diets 25LD, 50LD, 75LD and 100LD) as the primary protein source. The 100 lupin diet (LD) was formulated to include the maximum amount of sweet lupins according to the specifications for the specific species and the feeding phase. The maximum amount of lupins included in the 100LD therefore differs between the three feeding phases. The remaining three diets were formulated by mixing the diets to determine the gradual increase in lupins in the diets from 0LD up to 100LD. Soybean oilcake meal was thus gradually replaced by sweet lupins in the following ratios: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) and 0:100 (100LD) to make up the five dietary treatments for each feeding phase. No differences were found in the live weight of the birds at the end of each feeding phase or in the dry matter intake (DMI), average daily gain (ADG) or feed conversion ratio (FCR). The end weight (P = 0.07) and ADG (P = 0.09) for the starter phase tended to be higher for the birds on the 75LD. Birds fed the 50LD and 75LD tended (P = 0.08) to have the heaviest cold carcass weights, although dressing percentages did not differ. Birds receiving the 50LD diet had somewhat heavier (P = 0.05) thighs than those on the other diets. No differences were found for the weight of the big drum muscles of the birds. It was concluded that soybean oilcake meal can be replaced by sweet lupins in starter diets by up to 15% lupin inclusion in the diet (75LD) and in grower and finisher diets by up to 30% lupin inclusion in diet (100LD) without any significant detrimental effect on the production and slaughter traits.

The second study (chapter 4) evaluated the three primary ostrich products (leather, meat and feathers) to clarify whether the lupin inclusion levels had any effect on these products. Birds used in this study were the same birds as described in chapter 1. No differences were found for the marketable feather classes and measured leather traits, but the leather was thicker (P < 0.05) for

birds fed the 25LD. This corresponds with the heavier slaughter weight of the birds on the 25LD, although this was not significant. No differences were found in the chemical composition of the meat, apart from a higher ($P < 0.05$) intra-muscular fat content being found for birds on the 50LD. It can be concluded that the sweet lupin inclusion levels evaluated in this study had little influence on the leather traits, chemical composition of the meat of the birds measured (ten birds per treatment were selected around the median for chemical analysis of their meat) and the feather classes.

In the third study (chapter 5), 60 South African Black growing ostriches were randomly divided into 10 paddocks with six birds per paddock. Three trials with five experimental diets per trial were conducted to investigate the effect of sweet and/or bitter lupins on the feed preference of growing ostriches in a free-choice system. In Trial 1 (sweet) and Trial 2 (bitter), lupins replaced soybean oilcake meal in a step-wise manner for inclusion levels of 0%, 7.5%, 15%, 22.5% and 30%. In Trial 3, the soybean oilcake meal was replaced with 0%, 15% sweet, 15% bitter, 30% sweet and 30% bitter lupin inclusion levels. The position of the feeders containing each diet in successive paddocks changed by rotating the five feed troughs in a clockwise direction, but within each paddock the specific position of each feeder and diet stayed the same throughout the three trials. No interaction was found between day and diet for the three trials. The DMI per diet did not differ between the five treatments in any of the three trials. However, in trial 2 the birds showed a tendency ($P = 0.11$) to prefer the 7.5% bitter lupin diet to the 15% and 30% diets. The results showed that soybean oilcake meal can be replaced in the diets of growing ostriches by sweet lupin inclusion of up to 30% without any significant detrimental effect on feed preference and intake. The tendency ($P = 0.11$) of the birds to discriminate to some extend against the 15% and 30% bitter lupin diets may warrant further research.

This study found that lupins can be used without compromising growth or product quality in ostriches, making them a viable economic alternative.

Opsomming

Een van die grootste uitgawes van ‘n volstruisproduksie-eenheid is die voedingskostes wat ongeveer 75% tot die totale insetkostes bydra. Energie en proteïen is die twee belangrikste en volopste voedingstowwe in 'n gebalanseerde volstruisrantsoen en tussen die twee is die proteïenkomponent die duurste per eenheid gewig. Hierdie studie is dus onderneem om die insluiting van lupiene (*Lupinus angustifolius*) as ‘n alternatiewe proteïenbron in volstruisrantstoene (*Struthio camelus* var. *domesticus*) te evalueer.

In die eerste studie (hoofstuk 3) was daar vyf diëte met drie herhalings van tussen 15 – 17 kuikens elk. Die kuikens het almal dieselfde voor-aanvangs dieët ontvang. Vyf diëte is daarna geformuleer met behulp van ‘n rekenaargebaseerde program (Mixit2+) vir elke produksiestadium (aanvangs, groei en afronding) na aanleiding van spesifikasies soos uiteen gesit vir elk van die verskillende voedingsfases. Binne elke voedingsfase het die diëte of sojaboon oliekoekmeel (kontrole dieët, 0LD) of soetlupiene (alternatiewe proteïenbron, diëte 25LD, 50LD, 75LD en 100LD) as die primêre proteïenbron bevat. Die 100 lupien dieët (LD) was geformuleer om die maksimum hoeveelheid soetlupiene in te sluit, na aanleiding van die spesifikasies vir die spesifieke spesie en voedingsfase. Die maksimum hoeveelheid lupiene wat in die 100LD ingesluit is verskil dus tussen die drie voedingsfases. Die oorblywende drie diëte was geformuleer deur die diëte te meng om sodoende die geleidelike toename in lupiene van die diëte vanaf 0LD tot en met die 100LD te bepaal. Sojaboon oliekoekmeel was dus geleidelik vervang met soetlupiene in die volgende verhoudings: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) en 0:100 (100LD) om sodiende die vyf dieëtbehandelings vir elke voedingsfase te bepaal. Geen verskille is gevind in die lewendige gewig van die voëls aan die einde van elke voedingfase of in die droë materiaal inname (DMI), gemiddelde daaglikse toename (GDT) of voeromsetverhouding (VOV) nie. Die eindmassa (P = 0.07) en GDT (P = 0.09) vir die aanvangsfase het geneig om hoër te wees vir die voëls wat die 75LD ontvang het. Die voëls wat van die 50LD en 75LD ontvang het, het geneig (P = 0.08) om die swaarste koue karkasgewig te hê, alhoewel die uitslagpersentasie nie verskil het nie. Die voëls wat die 50LD ontvang het, het iewat (P = 0.05) swaarder dye gehad as die voëls op die ander diëte. Daar was ook geen verskille vir die gewig van die *Muscularis gastrocnemius* spiere van die voëls gevind nie. Daar is tot die gevolgtrekking gekom dat aanvangsdiëte tot en met 15% (75LD) soetlupiene kan bevat en groei- en afrondingsdiëte onderskeidelik tot en met 30% (100LD) soetlupiene kan bevat sonder enige betekenisvolle nadelige uitwerking op die produksie- en slageienskappe.

Tydens die tweede studie (hoofstuk 4) is die drie primêre volstruisprodukte (leer, vleis en vere), wat vanaf die volstruise in die eerste studie (hoofstuk 1) geoes is, geëvalueer om te bepaal of die lupien-insluitingsvlakke hierdie eind-produkte geaffekteer het. Geen verskille is vir die

bemarkbare vereklasse en vir die leereienskappe wat geëvalueer is gevind nie, maar die leer van die voëls wat die 25LD ontvang het was dikker ($P < 0.05$). Hierdie bevinding is in ooreenstemming met die swaarder slaggewig van die voëls wat die 25LD ontvang het, alhoewel dit nie betekenisvol was nie. Geen verskille is vir die chemiese samestelling van die vleis gevind nie, buiten vir die hoër binnespierse ($P < 0.05$) vetinhoud wat gevind is by die voëls wat die 50LD ontvang het. Ter opsomming is daar gevind dat die insluitingsvlakke van soetlupiene wat in hierdie studie getoets is ‘n baie klein invloed op die leereienskappe, chemiese samestelling van die vleis van die geselekteerde voëls (tien voëls per behandeling is rondom die median geselekteer vir verdere chemiese analises van die vleis) en die onderskeie vereklasse gehad het.

In die derde studie (hoofstuk 5) is 60 Suid-Afrikaanse volstruise lukraak in 10 kampe met ses voëls per kamp ingedeel. Drie proewe met vyf eksperiementele diëte per proef is uitgevoer om die voorkeur van groeiende volstruise ten opsigte van soet- en/of bitterlupiene in ‘n vrye-keuse stelsel te evalueer. In die eerste (soet) en tweede (bitter) proef het lupien-bevattende diëte sojaboon oliekoekmeel diëte met die volgende insluitingsvlakke in ‘n stapsgewyse manier vervang: 0%, 7.5%, 15%, 22.5% en 30% lupiene in die dieët. Tydens die derde proef, was die sojaboon oliekoekmeel verplaas met 0%, 15% soet, 15% bitter, 30% soet en 30% bitterlupien-insluitingsvlakke in die dieët. Die posisie van die voerbakke wat elke dieët bevat het, het in die opeenvolgende kampe van mekaar verskil deur die volgorde van die vyf diëte in ‘n kolksgewyse rigting te roteer, maar binne elke kamp het die spesifieke posisie van elke voerbak en dieët dieselfde gebly vir al drie proewe. Daar was geen interaksie tussen dag en dieët vir al drie die proewe gevind nie. Die DMI per dieët het nie verskil tussen die vyf behandelings in enige van die drie proewe nie. Tydens die tweede proef het die voëls egter ‘n neging ($P = 0.11$) getoon om ‘n voorkeur te hê vir die 7.5% bitterlupien-dieët en het tot ‘n mate gediskrimineer teen die 15% en 30% diëte met bitterlupiene. Vanuit die resultate kan daar tot die gevolgtrekking gekom word dat sojaboon oliekoekmeel in die diëte van groeivolstruise vervang kan word met soetlupien-insluitingsvlakke tot en met 30% sonder enige betekenisvolle nadelige effek op dieët-voorkeur en voerinname. Die neiging ($P = 0.11$) van voëls om teen die 15% en 30% bitterlupien-diëte te diskrimineer behoort verder ondersoek te word.

Hierdie studie het dus bevind dat lupiene gebruik kan word in volstruisdiëte sonder om die groeipotensiaal of kwaliteit van enige van die drie volstruis-eindprodukte nadelig te beïnvloed, wat lupiene ‘n lewensvatbare ekonomiese alternatiewe proteïenbron in volstruisdiëte maak.

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Notes

The language and referencing style used in this thesis are in accordance with the requirements of British Poultry Science. This thesis presents a compilation of manuscripts where each chapter is an individual entity and some repetition between chapters was therefore unavoidable. It should be known that each chapter has its own reference list instead of one comprehensive list appearing at the end of the thesis.

The following parts arising from this thesis have been presented at the following conferences and symposiums:

1. **The 48th South African Society for Animal Science Congress (SASAS), 21-23 September, Empangeni, Kwa-Zulu Natal, South Africa**

   J.A. ENGELBRECHT, T.S. BRAND & L.C. HOFFMAN (2015) Preliminary results on the utilisation of sweet lupins by ostriches during the starter phase. *Proceedings of the 48th South African Journal of Animal Science Congress,* Empangeni, South Africa, pp. 36 (Poster).

2. **The 6th International Ratite Symposium, 6-7 July 2016, Stellenbosch, Western Cape, South Africa**

   J.A. ENGELBRECHT, T.S. BRAND & L.C. HOFFMAN (2016) The evaluation of lupins (*Lupinus angustifolius*) as alternative protein source in ostrich diets. *Proceedings of the 6th International Ratite Symposium*, Stellenbosch, South Africa, abstract nr. 216 (Presentation).

   J.A. ENGELBRECHT, T.S. BRAND, L.C. HOFFMAN & A. ENGELBRECHT (2016) Feed preference of grower ostriches consuming diets differing in *Lupinus angustifolius* inclusion levels. *Proceedings of the 6th International Ratite Symposium*, Stellenbosch, South Africa, abstract nr. 99 (Presentation).

3. **The 5th Mediterranean Poultry Summit, 20-23 October 2016, Barcelona, Spain, Europe**

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List of abbreviations

ADF	Acid detergent fibre
ADG	Average daily gain
AFMA	Animal Feed Manufacturers Association
ANOVA	Analysis of variance
AOAC	Association of Official Analytical Chemists
BFMA	Balanced Feed Manufacturers Association
BSE	*Bovine spongiform encephalopathy*
BW	Bird weight
CF	Crude fibre
CP	Crude protein
DAFF	Department: Agriculture, Forestry and Fisheries
DM	Dry matter
DMI	Dry matter intake
EE	Ether extract (crude fat)
EU	European Union
EW	End weight
FCR	Feed conversion ratio
GLM	General linear model
HCl	Hydrogen chloride
HPAI	Highly pathogenic *Avian influenza*
HPLC	High pressure liquid chromatography
ICP	Inductively Coupled Plasma
IVOMD	*In vitro* organic matter digestibility
KKI	Klein Karoo International
LD	Lupin diet
LSMEANS	Least square means
ME	Metabolisable energy
MDC	Methylene dichloride
MeOH	Methanol
NAMC	National Agricultural Marketing Council
NDF	Neutral detergent fibre
NIRS	Near infrared reflectance spectrometer
NSPs	Non-starch polysaccharides
PVDF	Hydrophylic polyvinylidene difluoride syringe filter

SCOT	Southern Cape Ostrich Tanning
SE	Standard error
SOM	Soybean oilcake meal
TME	True metabolisable energy
TME_n	Nitrogen-corrected true metabolisable energy
TSA	Total sulphur containing amino acids

CHAPTER 1

GENERAL INTRODUCTION

Commercial ostrich farming started in South Africa around 1863, with the industry originally founded on the selection and breeding of ostriches for quality feather production for the export market (van Zyl, 2001; DAFF, 2013). Ostrich farming industries have also developed in Australia, northern Africa, Israel, Poland, Canada and the United States of America (Milton *et al*., 1994). Since the mid-1980s the commercial ostrich industry has grown rapidly in South Africa as a result of the increased demand for ostrich leather and consumers seeking a healthier alternative red meat to beef (Brand and Jordaan, 2011). However, the industry is characterised by a cyclic nature, with the relative values of the three products – feathers, leather and meat – varying depending on market trends and input costs. Currently the value of a slaughter bird is broken down into 45% each for the leather and meat and 10% for the feathers.

South Africa is regarded as the world leader in ostrich production, contributing *ca*. 75 – 85% of the global market share (DAFF, 2014; Kleyn, P., Pers. Comm., South African Ostrich Business Chamber, PO Box 952, Oudtshoorn, 6620, South Africa, 15 Sept. 2016). Oudtshoorn, a small town situated in the Klein Karoo region of the Western Cape Province of South Africa is regarded as the ostrich capital of the world, due to the high number of ostriches slaughtered from this district and the value-added products produced in this area (NAMC, 2010; DAFF, 2014). The South African ostrich industry contributes over R1 billion to the economy annually and currently employs over 50 000 people (Booysen, 2015).

The ostrich industry was severely impacted by the highly pathogenic *Avian influenza* (HPAI) H5N2 outbreak in April 2011 due to the resulting ban on the import of fresh ostrich meat into the European Union (EU). This caused a drastic decrease in the gross value of ostrich meat. The industry has since recovered to some extent, with the value of ostrich leather increasing, the development of pre-cooked (*sous vide*) meat products that were approved for export and an increase in the local consumption of ostrich meat. However, the lift of the four-year ban on the export of fresh ostrich meat into the EU in August 2015 should further enhance growth in the South African ostrich industry (DAFF, 2014; Booysen, 2015).

The continued challenges faced by the industry, such as the high costs of production in intensive systems, make the economical production of ostrich meat, leather and feathers difficult. In order to maintain an acceptable profit margin it is therefore important to optimise the aspects of a production system which can be controlled by the ostrich producers, such as nutrition. In intensive production units *ca.* 75% of the input costs are contributed by feed costs, necessitating the

identification of cheaper alternative raw materials to ensure the cost efficient production of slaughter ostriches. However, these alternatives must also be able to maintain optimum growth performance (Brand and Jordaan, 2004).

A large part of the nutritional costs are represented by the protein source, with expensive soybean oilcake meal being the major source of protein used in monogastric feeds throughout the world (Dalle Zotte *et al*., 2013; Snyman, 2016). It is therefore of vital importance to fully exploit the nutritional potential of all protein feeds that can be cultivated under local climatic conditions. Over the years, lupins (*Lupinus angustifolius*) have been identified as a potential alternative, locally-produced plant protein source which could be fed to animals at a positive profit margin and replace soybean oilcake meal as a raw material in ostrich diets. Concerns have been raised regarding the alkaloid content of lupin seeds; alkaloids are bitter tasting compounds, reducing the palatability of the feed. However, there are sweet (low in alkaloids, < 0.1%) and bitter (alkaloid-rich, 0.1 - 4%) varieties within the species (Breytenbach, 2005). During the 1950s and 1960s researchers in Western Australia concentrated on improving the agronomic characteristics and overcoming the bitter flavour of lupins (Gladstones, 1982), and as a result, more palatable varieties within *Lupinus angustifolius* were successfully cultivated. These are frequently referred to as Australian sweet lupin, which distinguishes it from bitter varieties grown elsewhere (Petterson and Fairbrother, 1996). However, it is important to remember that lupins can still only be included up to specific maximum levels to be utilised efficiently and to prevent undesirable effects (McDonald *et al*., 2011).

There is little literature available on the inclusion of lupins in ostrich diets and how dietary inclusion may affect the production and quality of the meat, leather and feathers produced by slaughter birds. However, the use of locally produced feed sources will benefit both the local grain legume industry as well as the ostrich industry and the potential of these sources is thus worth investigating. In an attempt to decrease the feeding costs of ostriches, least-cost diet formulations are applied by animal nutritionists and the use of lupins as a raw material can contribute to these diet formulations.

The aim of this study was therefore to investigate the nutritional potential of lupins (sweet and bitter) in ostrich diets by evaluating the effects of the treatment diets on the production traits, such as the dry matter intake (DMI), average daily gain (ADG) and feed conversion ratio (FCR). The quality of the three ostrich end-products were also evaluated in an attempt to assess the possible advantages or disadvantages of lupins as an alternative protein source in ostrich diets. In addition, the results of this study will contribute to the verification of the mathematical optimisation model for ostriches, designed by Professor Robert Gous of the University of Kwa-Zulu Natal and Professor Tertius Brand of the Western Cape Department of Agriculture. This model is used to

predict the effect of altering the diet on the ostrich end products and to formulate feeds to more precisely match the needs of the birds (Gous and Brand, 2008). Findings from this study will also broaden the knowledge of ostrich nutrition and aid in the formulation of least-cost diets that will provide for the needs of the birds at the specific growth stages (pre-starter, starter, grower, finisher and maintenance) without over- or undersupplying dietary nutrients.

The results of this study will also contribute to the identification of alternative feedstuffs that can be used in ostrich diets while optimising the cost-efficient production of slaughter birds. Furthermore, it is essential that all scientific information on the ostrich industry be utilised in the best interest of the industry to ensure its cost-competitiveness and liveability for future generations.

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CHAPTER 2

LITERATURE REVIEW

2.1. INTRODUCTION

Birds can be classified either as *Carinatae* or as *Ratite*. Ostriches form part of the ratite group, which are flightless or running birds that can cover ground at incredible speeds and are generally larger than flying birds (Strydom, 2007). The Emu (*Dromaius novaehollandiae*; Australia), Rhea (*Rhea Americana*; South America), Cassowary (*Casuarius*; Australia) and Kiwi (*Apteryx*; New Zealand) also form part of the ratite group and occur mainly in the Southern hemisphere (Strydom, 2007; NAMC, 2010; Hoffman, 2012). Wild ostriches prefer short-grass plains and semi-desert environments as their habitat. They are very heat tolerant, well adapted to living in desert and arid regions and can survive without water for relatively long periods (Ullrey and Allen, 1996).

The South African Black ostrich (*Struthio camelus* var. *domesticus*) was originally produced by crossbreeding the North African ostrich (*Struthio camelus camelus*) and South African ostrich (*Struthio camelus australis*) (Swart *et al*., 1987), to develop a breed known for its high quality feathers and more docile temperament. More recently, the potential of the ostrich as a meat producing animal has gained interest, resulting in the domestication of two wild subspecies, namely the Red Necks (*Struthio camelus massaicus*) and Blue Necks (*Struthio camelus australis*), with these also being incorporated into breeding programmes aimed at increasing meat production (Horbañczuk *et al*., 1998).

The ostrich is a multi-purpose animal, providing feathers, leather and meat, with the current value of these products contributing 10%, 45% and 45% respectively to the revenue generated from a bird (DAFF, 2014). Originally, feathers were the main export product, but at present the leather and meat are more sought-after. However, the industry has a cyclic nature, with the relative values of these products varying depending on the market demand and input costs. Nonetheless, ostrich farming is an established industry throughout the world (Sales and Hayes, 1996), with South Africa having *ca*. 75 – 85% of the global market share (DAFF, 2014; Kleyn, P., Pers. Comm., South African Ostrich Business Chamber, PO Box 952, Oudtshoorn, 6620, South Africa, 15 Sept. 2016) The highest concentration of ostriches in South Africa is found in the Western Cape Province in the Klein Karoo and Southern Cape regions (DAFF, 2014).

The raw materials needed to feed slaughter ostriches in an intensive ostrich production unit (feedlot) make up the largest proportion of the input costs (*ca*. 75%) (Jordaan *et al*., 2008), with Dalle Zotte *et al*. (2013) noting that the protein source represents a large part of these costs. Oilseeds such as soybean, sunflower and cotton are common plant protein sources that are included

in animal feeds, with soy being the most popular. The oilseed components are primarily cultivated in the summer rainfall areas of South Africa or need to be imported. Consequently the cost of transportation, exchange rates and market trends have a large influence on the price of these raw materials (NAMC, 2010). Alternative protein sources, such as lupins (*Lupinus angustifolius*), that can be cultivated locally and at a lower cost and do not affect the growth performance of the birds are therefore needed to ensure the cost-effective production of slaughter ostriches. The seed and vegetative parts of lupins are a rich source of protein and energy, but the fibrous seed coat and neutral detergent fibre (NDF) content of lupins will affect the digestibility of the meal. However, the digestive system of the ostrich enables them to digest large amounts of fibre-rich plant material (Brand *et al*., 2002), thus making lupins a potentially valuable alternative protein source which could allow the formulation of cheaper diets.

Although research on the protein and energy requirements of ostriches has increased in recent years, research evaluating alternative protein sources such as lupins is still limited. Therefore this review aims to examine the potential of lupins as a possible alternative, locally produced, plant protein source in the diets of slaughter ostriches and to apply these findings in the best interest of the industry.

2.2. THE OSTRICH INDUSTRY

Since the establishment of the ostrich industry, ostrich production has transformed into an intensively managed farming activity and become an important part of the agricultural industry (Jordaan *et al*., 2008; WCDA, 2014). Over the past 10 years, the average annual gross value of ostrich production amounted to about R1.5 billion (DAFF, 2014). However, in comparison with other livestock species, the ostrich industry is still relatively new, as the species has not been domesticated for as long as traditional livestock species (Brand and Olivier, 2011; Cloete *et al*., 2012).

2.2.1. History of the ostrich industry

Commercial ostrich farming started around 1863, when a few ostrich farmers in the Karoo and Eastern Cape managed to tame ostriches and breed them in captivity to produce feathers for export to Europe on a large-scale (van Zyl, 2001; DAFF, 2013). During the early 1900s the South African government and farmers launched a cross-breeding programme to develop a breed with plumes of a higher quality for the international feather market, as well as a docile nature for easier handling (Strydom, 2007). This selection for a smaller, tamer bird resulted in the development of the South African Black ostrich (*Struthio camelus* var. *domesticus*) through crossbreeding the North African ostrich (*Struthio camelus camelus*) and the South African ostrich (*Struthio camelus*

australis) (Swart *et al*., 1987). With the shift of income from feathers to meat and skins larger birds have become more favoured, leading to the incorporation of two wild subspecies, namely the Red Necks (*Struthio camelus massaicus*) and Blue Necks (*Struthio camelus australis*) into breeding programmes for increased meat production (Horbañczuk *et al*., 1998). However, the South African Black is still the most common breed in ostrich husbandry systems.

Ostrich feathers were the fourth largest export product from South Africa in 1913 after gold, diamonds and wool, but soon thereafter the industry collapsed as a result of changes in fashion and the First World War (DAFF, 2013; Jorgensen, 2014). A resurge was experienced after the Second World War, with the market expanding to include not only feathers but also leather and tourism as new sources of income (Anon, 2004). In 1970, the first leather tannery for ostrich skins was built, and ostrich leather currently remains a prime source of income for ostrich farmers (NAMC, 2010). The first ostrich abattoir was opened in 1964 (Jorgensen, 2014), and the first abattoir for the export of ostrich meat to Europe in 1993 (DAFF, 2013). The emphasis shifted to meat production during the early 1990's, with ostrich meat entering the international market as a specialty meat product, due to its health benefits as a rich source of protein and iron and low fat and cholesterol content (NAMC, 2010; DAFF, 2013). The outbreak of *Bovine spongiform encephalopathy* (BSE) and *Foot-and-mouth* disease in large parts of Europe in 2000 caused the production of ostrich meat and the price thereof to increase steeply. This was as a result of the increased demand by consumers wanting to eat healthier meat and seeking a safe alternative red meat to beef (Brand and Jordaan, 2011).

2.2.2. Ostrich production systems

Ostriches can be reared extensively, semi-intensively and intensively. In an extensive production system, the birds are exclusively dependent on the natural grazing and/or cultivated pasture, such as lucerne, canola and salt-bush plantations (Brand, 2014). Although this is the cheapest form of feed for animals it is important to maintain the correct stocking rate, as it is well known that foraging ostriches can destroy natural veld when the stocking density is too high (Strydom *et al.*, 2009). Brand (2014) found that irrigated lucerne pasture to have a carrying capacity of 10 birds per hectare (ha). Furthermore, the natural feeding behaviour of foraging ostriches may have a destructive influence on grazing, due to trampling or stripping leaves from the stem of the plant (Brand, 2014). However, in order to obtain efficient growth and optimal weight gain, it is advised to supplement natural grazing and/or cultivated pasture to prevent any potential nutrient deficiencies (Strydom *et al.*, 2009; Brand, 2014).

A semi-intensive production system retains natural veld or cultivated pasture as a primary feed source but includes the provision of a concentrate to supplement any nutritional deficiencies.

The last form of production system is the intensive production system, where the birds receive a nutritionally balanced feed ration and have no access to natural grazing and/or cultivated pasture. In South Africa, this is the most common practice. The birds are generally moved to an intensive feeding system after being raised on cultivated lucerne pastures from two weeks of age to about three to four months old (Brand, 2014).

2.2.3. Ostrich end products and economic potential

The ostrich was initially developed as a single-product animal, with the identity of the primary product being determined by cyclic market changes. However, in recent times full utilisation of the multi-product nature of the ostrich has become an economic necessity (Adams and Revell, 2003). The South African ostrich industry is thus characterised by three distinct products, feathers, leather and meat, and currently the value of a slaughtered bird for these products is broken down into 10%, 45% and 45% respectively (NAMC, 2010; DAFF, 2014).

There is an international market for ostrich products, with South Africa being regarded as the world leader contributing about 75 – 85% of the global production (DAFF, 2014; Kleyn, P., Pers. Comm., South African Ostrich Business Chamber, PO Box 952, Oudtshoorn, 6620, South Africa, 15 Sept. 2016). Ostrich feathers are not only a popular fashion product and used for carnivals, but are also used to make industrial and household dusters (van Zyl, 2001; DAFF, 2014). Ostrich leather is regarded as a luxury product and the presence of the feather follicles or nodules on the leather gives it its unique appearance. It is one of the most attractive, supple and durable leathers and the nodule diameter and shape contributes to the quality of the leather (Meyer *et al*., 2004; NAMC, 2010; Engelbrecht, 2014). The potential value of ostrich meat was only recently appreciated, with its health benefits including a low sodium content, favourable fatty acid profile, low intra-muscular fat and cholesterol contents and high iron and vitamin E contents (Mellett, 1992; Sales and Oliver-Lyons, 1996; Majewska *et al*., 2009; Poławska *et al*., 2011).

Ostriches are generally slaughtered at 10 – 14 months of age and produce approximately 27 kg of meat, 4.2 m^2 of leather and 1 kg of feathers (DAFF, 2014). Experimental information on the production of the three ostrich end products at the different slaughter weights are presented in Table 2.1. Decision-making with regard to the ideal slaughter age depends on the market demand/consumer preference for specific features in the end products and the volatile feed prices, which greatly influence the profitability of a production system. Therefore the ideal slaughter age of an ostrich may not always be the same, depending on the specific market trends (Brand, 2014).

The ostrich industry was severely impacted by the highly pathogenic *Avian influenza* (HPAI) H5N2 outbreak in April 2011. As a result of this outbreak, the European Union (EU) banned the import of fresh meat and a drastic decrease in the gross value of ostrich meat was experienced.

However, the industry recovered to some extent by increasing the value of ostrich leather, successfully developing pre-cooked (*sous vide*) meat products that could be exported and increasing the local consumption of ostrich meat (DAFF, 2014; Booysen, 2015). In August 2015, the four-year ban on the export of fresh ostrich meat to the EU was lifted, which has contributed to the increasing growth of the South African ostrich industry, which currently employs over 50 000 people (Booysen, 2015).

Table 2.1. *Experimental information on the production of the three ostrich end products per slaughter bird at different slaughter weights (Brand, 2014)*

Slaughter weight (kg)	Meat			Leather			Feathers
	Carcass mass (kg)	Dressing percentage (%)	Total high-price meat cut yield per carcass (kg)	Skin surface (dm^2)	Follicle size (mm)	Average grade (1 - 5)	Feather yield (kg)
65	31	48	14.2	127	3.03	1.68	1.12
72	34	47	15.2	130	3.10	1.81	1.20
79	37	47	16.2	134	3.17	1.94	1.28
86	39	45	17.2	138	3.24	2.07	1.36
94	42	45	18.2	141	3.31	2.20	1.44
101	45	45	19.2	146	3.38	2.33	1.52
107	47	44	20.1	149	3.45	2.46	1.60
115	50	44	21.1	153	3.52	2.59	1.68
122	52	43	22.1	157	3.59	2.72	1.76

2.2.4. Ostrich production areas

The ostrich industry is predominantly South African, with the highest concentration (*ca.* 75%) of ostriches being found in the Western Cape Province in the Klein Karoo and Southern Cape regions (Figure 2.1.) (Brand, 2007; DAFF, 2014). Ostriches do have a relatively open breeding season and breed throughout the year. In the southern hemisphere peak production for ostriches occurs between winter (July) and summer (December). Thus, the onset of their peak breeding season falls during winter and proceed till late spring/early summer. With modern farming and breeding practices it is possible to manipulate the breeding season by using periods of rest (separation of male and female flocks), nutrition and feeding practices (flush feeding) (Lambrechts, 2004). Ostriches lay their eggs in open nests and the presence of water will cause the eggs to rot. Furthermore, ostrich chicks are very vulnerable and wet circumstances will not favour the survival

of the offspring (Lambrechts, 2014). Therefore the western, drier part of South Africa, which is a winter rainfall region, is more suitable for ostrich farming (NAMC, 2010).

Oudtshoorn is situated in the Klein Karoo and is known as the ostrich capital of the world, due to the high number of ostriches slaughtered from this district and the value-added products produced in this area (NAMC, 2010; DAFF, 2014). However, ostrich production systems are found throughout South Africa (except in KwaZulu-Natal) on about 588 registered export farms, of which 453 farms are in the Western Cape, 102 are in the Eastern Cape and 33 are in the rest of the country. The Eastern Cape contributes about 19% of the national ostrich flock, while the remaining 6% are found in the Free State, Limpopo and Northern Cape (Figure 2.1.) (DAFF, 2014).

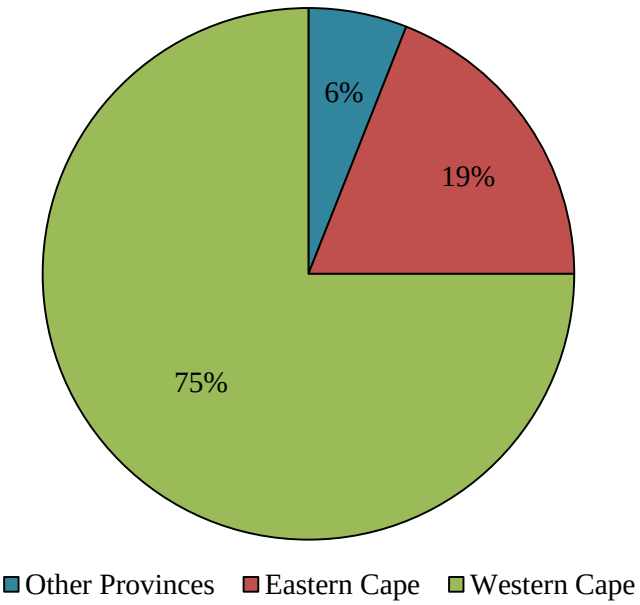

Figure 2.1. *Ostrich production areas in South Africa (DAFF, 2014)*

2.2.5. Challenges of an ostrich enterprise

On the farm level, feed costs are known to be the largest cost factor in most livestock production systems. In an intensive ostrich production unit, nutrition contributes approximately 75% of the total input costs to produce a bird that is ready for slaughter (Brand and Jordaan, 2011), with protein sources representing a large proportion of this (Dalle Zotte *et al*., 2013). An increase in the price of traditional protein sources, such as soybean oilcake meal, necessitates the development of cheaper alternatives to ensure the cost efficient production of slaughter ostriches without affecting their growth performance and end products. However, reliable scientific information regarding the nutritional requirements of ostriches, the nutritive value of raw materials and nutritional management systems is limited. Any increase in reliable scientific information will enhance the scope of opportunities and the profitability of the industry.

To ensure the viability and profitability of the ostrich industry for future generations, further research is also necessary in the fields of chick survival (mortality rate 30 – 40%) and ratite behaviour. This will contribute to the knowledge needed to run a commercial ostrich enterprise as efficiently and sustainably as possible while simultaneously improving the liveability and welfare of the birds (Brand and Gous, 2006; Cloete *et al*., 2012). Since ostrich farming is still a relatively new industry, reliable scientific information is limited relative to that available for other livestock species.

2.3. OSTRICH DIGESTION AND NUTRITION

Dowsley and Gardiner published the first book on ostrich nutrition in 1913 (Cloete *et al*., 2012), with pioneering research and comprehensive reviews subsequently being produced by various researchers (Swart, 1988; Du Preez, 1991; Cilliers, 1995; Angel, 1996; Cilliers and Angel, 1999; Brand and Gous, 2006; Gous and Brand, 2008; Brand and Olivier, 2011, to name but a few). However, knowledge of ostrich nutrition is still in its infancy relative to that of other monogastric animals.

Models (EFG Software 2008) developed by Gous and Brand (2008) to simulate the feed intake and growth of poultry and pigs are now also being applied to ostriches, in the hope of improving the competitiveness of the ostrich industry in terms of production and feed manufacturing (Brand, 2007; Gous and Brand, 2008). These mathematical optimisation models take into account growth, production, environmental and feed factors (including price), as well as the interaction between these factors. This results in the production of a least-cost diet formulation which meets the nutritional requirements of the birds at a specific developmental stage and allows the production of end products of a specific quality as required by the market (Brand, 2007). The optimisation model for ostriches is more complicated than for pigs, for example, where meat is the only product, because all three end products need to be taken into account.

2.3.1. Anatomy of the digestive system

The digestive tract of the ostrich, similar to that of other birds, consists of a beak, mouth, oesophagus, proventriculus (glandular stomach), gizzard (muscular stomach), small intestine (duodenum, jejenum and ileum), large intestine (two caeca and the proximal, middle and distal colon or rectum) and cloaca (Figure 2.2.). Ostriches are monogastric herbivores and are distinguished from other monogastric species by their relatively large digestive tract, which conveys the ability to digest fibrous plant materials (Brand and Gous, 2006; Brand and Olivier, 2011).

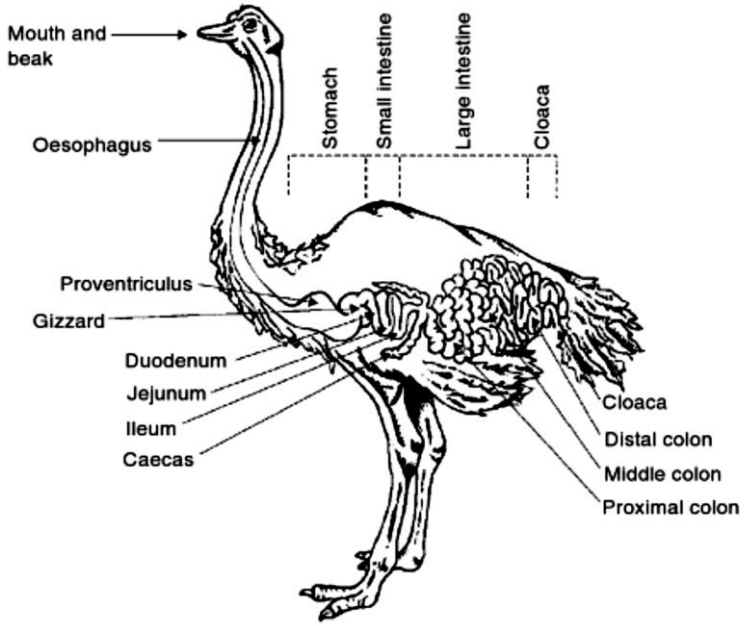

Figure 2.2. *Graphic illustration of the digestive system of the ostrich (Brand and Gous, 2006)*

The digestive process starts in the proventriculus, where hydrochloric acid and enzymes are secreted (Brand and Gous, 2006). Thereafter the ingested plant material or feed is mechanically ground to a finer form in the gizzard, through the contractions of the muscular gizzard wall and abrasion by stones and pebbles that are also ingested by the bird to facilitate digestion (Brand and Olivier, 2011). The partially digested feed is further digested in the alkaline environment of the small intestine by various digestive enzymes (Iji *et al*., 2003) (amylase, trypsin, chymotrypsin, lipase, maltase, sucrose, alkaline phosphatase and arginase), where the absorption of digested nutrients also takes place. However, ostriches lack the enzyme cellulase to digest plant fibre components, and they thus rely on hindgut fermentation in the large intestine, together with a slow rate of passage of the digesta, to break down these components (Cilliers and Angel, 1999). Partially digested feed is exposed to microbial fermentation in the alkaline environment of the colon, enabling the digestion and utilisation of fibrous material (Brand and Gous, 2006).

Cilliers and Angel (1999) reported that ostriches are without any doubt the best hindgut fibre fermenters among birds and that their ability to digest plant fibre is mainly due to modifications to their hindgut. However, ostrich chicks only develop their ability to digest fibrous material after approximately three months of age (Angel, 1996), with the gastrointestinal tract developing from a typical bird neonate to that of a hindgut fermenter in a space of 70 – 80 days (Brand and Gous, 2006). This gives rise to the need for the different feeding phases at specific growth stages (pre-starter, starter, grower, finisher and maintenance) when ostriches are reared intensively. Swart (1988) initially described the ability of ostriches to utilise low quality raw materials and found that

ostriches effectively digest 66% of hemicellulose and 38% of cellulose. Furthermore, Swart (1988) also found that the ostrich could possibly obtain 12 – 76% of their energy in the form of volatile fatty acids, which are produced as end products of fibre digestion in the large intestine. Brand *et al*. (2000b) and Brand *et al*. (2002) found that in comparison to poultry and pigs, ostriches utilise 30% and 20% more energy respectively on the same feeds.

2.3.2. Nutritional requirements and production norms

According to Cilliers and Angel (1999) it is important to first understand the natural diet and physical and functional properties of the digestive tract, before trying to determine the nutritional requirements of any species. In addition, a sound knowledge of the nutrient composition of the raw materials and nutrient make-up of the bird during the various growth stages is necessary to better formulate a diet that is balanced and meets the needs of the bird at the various feeding phases (Cilliers and Angel, 1999). The under- or oversupply of certain nutrients can have a negative impact on the production performance and health of the bird and raw materials are wasted in the process, having negative cost implications (Brand *et al*., 2014).

In ostrich feeds, the following nutritional components are important and should be included in the diet: energy (carbohydrates and fat), protein (amino acids), minerals, trace elements and vitamins (Brand, 2014).

Energy

Scientific information on metabolisable energy (ME) values for ostriches is limited and therefore poultry energy values were used to formulate ostrich diets up to 1995 (Angel, 1996). Calorimetric studies by Swart *et al*. (1993) found the maintenance energy requirements for ostriches to be 0.44 MJ/kg $W^{0.75}$ per day and the efficiency of ME utilisation for growth (tissue synthesis) to be 0.32. Cilliers *et al*. (1994) and Cilliers (1995) determined the nitrogen-corrected true ME (TME_n) values of feeds for ostriches and concluded that the TME_n is a more accurate estimate of the energy value of the feed, as it is corrected for nitrogen retention. Brand *et al*. (2000b) found that ostriches obtained about 30% more TME_n than poultry from low energy, high fibre diets. This can be ascribed to the hindgut fermentation ability of the ostrich, enabling them to utilise high fibre diets and derive energy from the diets in the form of volatile fatty acids (Swart, 1988). Ostriches are thus far more efficient in their energy utilisation than poultry and therefore the obesity problems experienced in the ostrich industry were due to an oversupply of energy in the diets (Angel, 1996).

Dietary feed intake in the ostrich is determined by the energy content of the feed; therefore, as the energy density of the feed increases, a decrease in feed intake will be observed (Brand *et al*., 2000a; Brand *et al*., 2000b). The average feed intake and feed energy values of the different ostrich

growth phases (Table 2.2) indicate that as the bird ages, the energy requirements decrease. This is due to the high energy demands of the rapid muscle and bone growth in young animals, whereas mature birds only require energy for maintenance. Furthermore, as the physiological state of the ostrich gastrointestinal tract progresses toward a hindgut fermenter, its ability to utilise fibre to produce energy in the form of volatile fatty acids, also increases (Swart 1988).

Table 2.2. *Average feed intake and feed energy values of the different ostrich feeding phases (adapted from Brand and Gous (2006) and Brand and Olivier (2011)*

Feeding phases	Age (months)	Dry matter intake (g/bird/day)	TME* (MJ ME/kg feed)
Pre-starter	0 - 2	275	14.5
Starter	2 - 4.5	875	13.5
Grower	4.5 - 6.5	1603	11.5
Finisher	6.5 - 10.5	1915	9.5
Maintenance	10.5 - 12	2440	8.5

*TME – True metabolisable energy

Protein and amino acids

Amino acids, the basic units of proteins, are the building blocks for the synthesis of tissue components, enzymes, hormones and carriers, making protein a vital part of the diet. However, the amino acid profile of the diet is just as important, if not more so, than the total protein content. Any imbalances in the amino acid profile are often found to be costly, due to the consequent inefficient utilisation of the individual amino acids (Viviers, 2015) and the metabolic cost of deamination.

Carstens (2013) stated that protein concentrations of 28% and above can lead to physical deformities in ostriches and negatively impact the financial outcomes of an ostrich enterprise. Table 2.3 provides the predicted mean dry matter intake for each growth stage along with the corresponding protein and amino acid requirements.

Table 2.3. *Predicted mean dry matter intake with corresponding protein and amino acid requirements for ostriches at the different growth stages (adapted from Du Preez (1991), Du Preez et al. (1992), Cilliers et al. (1998) and Brand and Olivier (2011))*

Predicted parameter	Feeding phases				
	Pre-starter	Starter	Grower	Finisher	Maintenance
Live mass (kg)	0.85 - 10	10 - 40	40 - 60	60 - 90	90 - 120
Age (months)	0 - 2	2 - 5	5 - 7	7 - 10	10 -20
Feed intake (g/day)	275	875	1603	1915	2440
Crude protein (g/100 g feed)	22.9	19.7	14.7	12.2	6.92
Lysine (g/100 g feed)	1.10	1.02	0.84	0.79	0.58
Methionine (g/100 g feed)	0.33	0.33	0.29	0.28	0.24
Cystine (g/100 g feed)	0.23	0.22	0.18	0.17	0.14
Total SAA (g/100 g feed)	0.56	0.55	0.47	0.45	0.38
Threonine (g/100 g feed)	0.63	0.59	0.49	0.47	0.36
Arginine (g/100 g feed)	0.97	0.93	0.80	0.78	0.63
Leucine (g/100 g feed)	1.38	1.24	0.99	0.88	0.59
Isoleucine (g/100 g feed)	0.70	0.65	0.54	0.51	0.38
Valine (g/100 g feed)	0.74	0.69	0.57	0.53	0.36
Histidine (g/100 g feed)	0.40	0.43	0.40	0.40	0.37
Phenylalanine (g/100 g feed)	0.85	0.79	0.65	0.61	0.45
Tyrosine (g/100 g feed)	0.45	0.44	0.38	0.38	0.31
Phenylalanine and tyrosine (g/100 g feed)	1.30	1.23	1.03	0.99	0.76

Vitamins and minerals

There is currently very little information available on the vitamin and mineral requirements of ostriches, and data from other species is therefore used when ostrich feeds are formulated (Brand and Olivier, 2011; Brand, 2014). Law 36 of 1947 regulates the South African commercial guidelines for calcium and phosphate of ostrich feeds, as presented in Table 2.4.

Table 2.4. *Commercial guidelines for calcium and phosphate of ostrich feeds for the different feeding phases (adapted from Brand, 2014)*

Feeding phases	Min. calcium (g/kg)	Max. calcium (g/kg)	Min. phosphate (g/kg)
Pre-starter	12	15	6
Starter	12	15	6
Grower	10	16	5
Finisher	9	18	5
Maintenance	8	18	5

Production norms

The nutritional composition of the feed provided to growing ostriches will determine their growth rate (Brand and Gous, 2006). Table 2.5 contains practical production norms and the annual feed requirements per slaughter bird throughout the different feeding phases. The values in Table 2.5 are based on several studies by Brand and Jordaan (2004) for slaughter ostriches under intensive feedlot conditions and can serve as a guideline for a commercial ostrich enterprise.

Table 2.5. *Practical production norms and the annual feed requirements per slaughter bird (adapted from Brand and Jordaan (2004) and Brand and Gous (2006))*

Feeding phases	Age (months)	Live mass (kg)	DMI[1] (g/bird/day)	ADG[2] (g/bird/day)	FCR[3] (kg feed/kg gain)	Annual feed intake (kg/bird)*	Cumulative feed intake (kg/bird)*
Pre-starter	0 - 2	1 - 10	275	150	1.80	16	16
Starter	2 - 4.5	10 - 40	1100	400	2.75	84	100
Grower	4.5 - 6.5	40 - 60	1650	330	5.00	100	200
Finisher	6.5 - 10.5	60 - 90	2500	240	10.0	300	500
Maintenance	10.5 - 12	90 - 120	3000	200	15.0	150	650

[1]Dry matter intake

[2]Average daily gain

[3]Feed conversion ratio

*Based on feeding a pelleted ration

2.3.3. Raw materials

The feed conversion ratio and growth rate of an ostrich can be enhanced by replacing cheaper fibrous feed sources with a more nutrient-dense diet. Therefore the decision made on which feedstuffs to use as raw materials in ostrich diets will always be price-related and dependent on market trends (Brand and Olivier, 2011). However, when diets and feeding systems of ostriches are formulated, the hindgut fermentation ability of the ostrich should be taken into account to produce slaughter ostriches and their products in an economic manner (Brand and Olivier, 2011). Furthermore, Brand (2007) found that the use of optimisation models and least-cost diet formulations resulted in a 10% saving in feed costs. For the South African ostrich industry as a whole this would result in a saving of more than R55 million per annum.

The most important raw materials used in South Africa when formulating ostrich feeds are presented in Table 2.6. In South Africa, the most common method of ostrich production is under intensive feeding conditions with a balanced diet being provided for each specific feeding phase, in order to best meet the nutritional needs of the bird.

Table 2.6. *The most important raw materials used in ostrich feeds (Brand, 2014)*

Concentrates	Roughage	Protein sources	Mineral sources	Other
Maize	Lucerne hay	Soybean oilcake	Feed lime	Synthetic lysine
Barley	Wheat bran	Canola oilcake	Di-calcium phosphate	Synthetic methionine
Wheat	Oat bran	Sunflower oilcake	Mono-calcium phosphate	Plant oil
Triticale	Barley hay	Fish meal	Salt	Molasses products
Oats	Oat hay	Full-fat soy (roasted)	Mineral and vitamin premix	Binding agents
Brewer's grain	Oat straw	Full-fat canola		Medicines e.g.
	Wheat straw	Sunflower seeds		- antibiotics
	Silage	Sweet lupins		Additives e.g.
		Peas		- growth promoters
		Beans		- pre- and probiotics
		Gluten		- etc.

The balanced diets are manufactured using various raw materials, mixed in specific ratios as formulated by a registered animal nutritionist for the different feeding phases and should be presented to the birds in the physical form as recommended in Table 2.7 (Brand, 2014).

Table 2.7. *Recommended physical form of ostrich feeds for the different feeding phases (adapted from Brand, 2014)*

Feeding phases	Age (months)	Processing (sieve gauge)
Pre-starter	0 - 2	Meal
Starter	2 - 4.5	Crumbs
Grower	4.5 - 6.5	Pellets (6 mm - 8 mm)
Finisher	6.5 - 10.5	Pellets (6 mm - 8 mm)
Maintenance	10.5 - 12	Pellets (6 mm - 8 mm)

2.4. OVERVIEW OF THE SOUTH AFRICAN FEED INDUSTRY

The South African feed industry was established around 85 years ago, during the severe droughts and depression that occurred in the 1930's (DAFF, 2015; AFMA, 2016). Scientific research into the feeding of farm animals was necessitated by these circumstances and consequently alternative feeding systems were developed (AFMA, 2016). In 1945 the Balanced Feed Manufacturers Association (BFMA) was established after the need for a structured industry body to act on behalf of the industry and take care of the industry's interests was identified (AFMA, 2016). In 1988 the association's name was changed to the Animal Feed Manufacturers Association (AFMA), under whose guidance the South African feed industry has taken enormous leaps to ensure "safe feed for safe food" (Hofmeyr, 2013; AFMA, 2014; AFMA, 2016).

South Africa has developed a sophisticated and professional livestock industry which can compete with the best in the world. This is largely as a result of the stability and advancement of the commercial feed industry, which allows the nutritional needs of livestock to be met as well as the production of animal proteins at a competitive price and a profitable margin (Hofmeyr, 2013). South African livestock feed is produced by both companies belonging to AFMA and non-AFMA members and is based on the requirements of the livestock in the country (AFMA, 2014). The feed industry has expanded and become more industrialised in recent years to meet the higher demand of the livestock and poultry sectors. By feeding balanced, complete rations, the productivity of livestock has been improved and increased growth rates and better feed conversion efficiencies have been attained (Moran, 2016).

In 2014, South Africa produced 11.4 million tons of animal feed (AFMA, 2015). Therefore, in the overall food production process, animal feeds play an important role to ensure safe, abundant and affordable animal proteins. Least-cost diet formulation is also applied by animal nutritionists in an attempt to minimise feeding costs. However, it remains a challenge to the nutritionist to formulate a balanced diet that will ensure desirable production performance (Viviers, 2015).

2.4.1. Feed production and challenges - locally and worldwide

Hofmeyr (2013) noted that the animal feed industry of South Africa operates within an environment with unique challenges. Currently it is threatened by an increase in competitive intensity in direct relation to the growing overcapacity of the industry. Furthermore, the demand from South African consumers for all animal protein products, ranging from monogastrics to ruminants, has decreased due to the economic pressure experienced by the South African consumer. Rising living costs, which reached record levels in 2012, were responsible for this pressure. Nonetheless, feed industries that have focussed on increasing efficiency and controlling costs have ensured that they remain positioned to provide for the food sector in future generations (Hofmeyr, 2013). In 2012, international feed production figures amounted to 954 million tons (AFMA, 2014), with this showing a modest increase of 1% to 963 million tons during 2013. This relatively minimal increase in production is likely due to the droughts experienced in more than 30 countries worldwide in 2012 (AFMA, 2014).

The global feed industry was impacted by a number of factors throughout 2014. These included widespread droughts, which led to an increase in the price of raw materials, fluctuating import/export standards and animal diseases (*Avian influenza* and *Porcine epidemic diarrhea*), which were disastrous for many farmers. However, relative to 2013 international feed production increased 2% in 2014, reaching 980 million tons (AFMA, 2015). In contrast, the South African harvest was smaller than expected in 2015. This caused the prices of raw materials to be highly volatile and the reduced quantities available resulted in a substantial decline in the gross income from field crops. The poor harvest was due to the summer drought, primarily caused by the occurrence of an El Niño effect, which created various uncertainties in terms of weather conditions and planting intensities in the Southern hemisphere (AFMA, 2015; Snyman, 2016).

2.4.2. Analysing the global market share from a species perspective

In terms of the global market share of feed sales for each species, poultry feed accounts for the largest share, followed by pigs, ruminants, aquaculture, pets, equines and other species (AFMA, 2015; DAFF, 2015). The global feed production per species is presented in Figure 2.3 (AFMA, 2015).

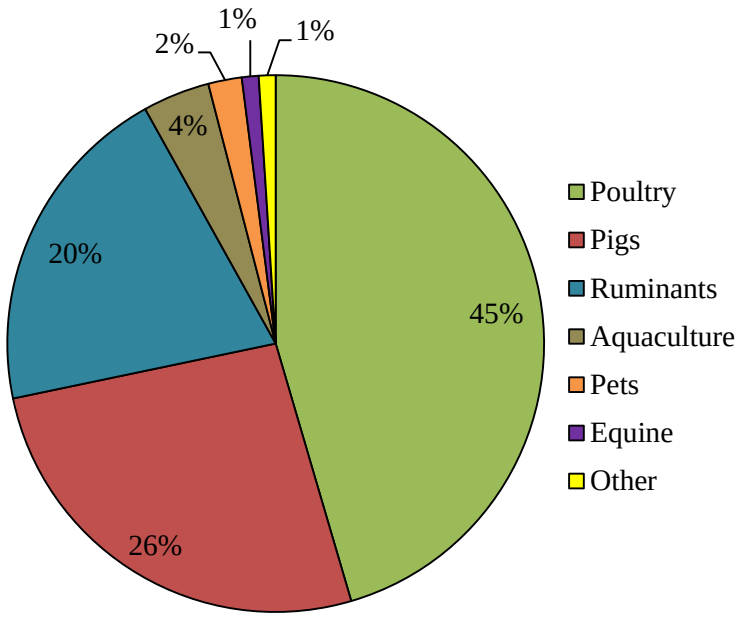

Figure 2.3. *Global feed production per species (million tons) (AFMA, 2015)*

Poultry held its position as the industry leader with 45% (439 million tons) of the feed market share, despite a 1.2% decrease in the poultry feed sector during 2013/2014. The dominance of the poultry industry is likely due to the high demand for good quality poultry products (meat and eggs), in part due to their affordable price, which is made possible by their efficiency of production (AFMA, 2015; DAFF, 2015). The highest growth was achieved by the pig feed sector, with an increase of 6.3% (258 million tons) (AFMA, 2015). Since 2011 feed production in this sector has increased gradually, despite raw materials becoming more expensive (Moran, 2016).

The dairy, beef and small ruminants feed market declined during 2013, but in 2014 there was an increase of 2.2%. High feed prices in times of drought and raw material scarcity forces producers to utilise alternative feed resources. The ruminant sector has the most alternative feed materials available to alleviate costs, such as natural grazing and fodder (AFMA, 2015; Moran, 2016).

In the aquaculture industry, a record growth of 17% was recorded for aquafeed during 2013. This growth continued in 2014, although at a slower pace, achieving 2.7% growth (AFMA, 2015). Equine feed and pet foods still remain low at 3% of the total feed tonnage produced, but they are likely to continue to show a gradual upward trend (Moran, 2016).

2.4.3. South Africa animal feed production segmental shares

The feed industry produces a variety of feeds to serve the needs of the various sectors, ranging from livestock to pets. These feeds are specifically formulated from highly nutritious raw materials to enhance the growth and development of the animals and improve their productivity. In order to produce compound feeds various agricultural raw materials are required. Therefore the main factors determining the composition of a nutritionally balanced diet includes the raw material prices, the nutritional value of the different feed components, the nutritional requirements of the particular animal and the government's rules and regulations (DAFF, 2015). In the South African feed industry, the majority of raw materials are imported (for example the United States of America, Zambia, Benin, United Kingdom, Malawi, Ethiopia, Argentine, Mozambique and Chile) and the compound feeds then are formulated and mixed locally.

The contributions of the five major animal feed categories (broilers, beef and sheep, dairy, layers and pigs), along with the smaller sectors, to the South African animal feed industry can be seen in Figure 2.4 (DAFF, 2015). In 2014, broiler feed accounted for approximately 30% of the animal feed production segmental shares, followed by 29% for beef and sheep. Globally pig feed production ranked second, but in South Africa only 7% of the total feed produced is for the pig sector. The remaining 5% is made up by the other animal feed sectors (DAFF, 2015).

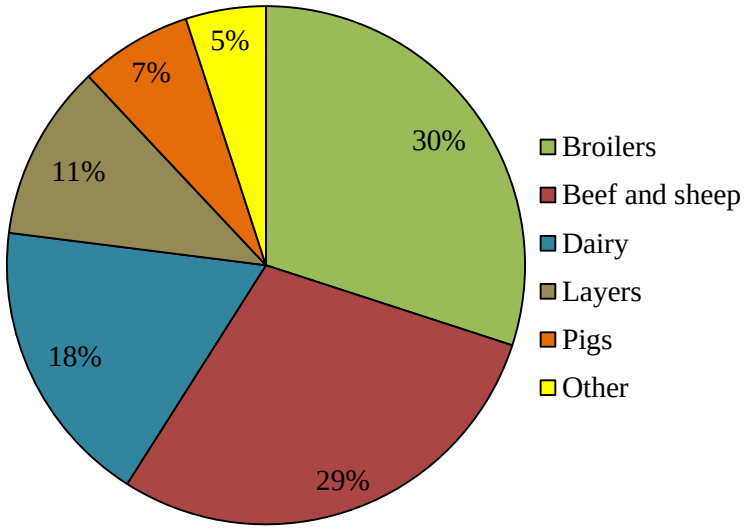

Figure 2.4. *South African animal feed production segmental shares (DAFF, 2015)*

2.4.4. The way forward

Compound feed production is fast approaching an estimated annual figure of 1 billion tons worldwide. In addition to this, about 300 million tons of feed are produced directly by on-farm mixing, which brings the global feed production to an estimated 1.3 billion tons annually (AFMA,

2015; DAFF, 2015). The feed industry is well-established and will grow in the future to provide for the animal protein needs of the consumer (Moran, 2016). However, it will continue to face a broad spectrum of challenges, such as food security and rising input costs, a world population expected to reach nine billion people around 2050 (Godfray *et al*., 2010; Hofmeyr, 2013), a decrease in available cropland and the more frequent occurrence of droughts (Hofmeyr, 2013; Moran, 2016).

2.5. THE CURRENT SITUATION OF PROTEIN RESOURCES IN SOUTH AFRICA

In animal nutrition, maize is considered as the main energy source and soybean oilcake meal the main protein source (Brandt, 1998). In South Africa the raw materials used to manufacture animal feeds are adequately available, with maize in particular being a major feed ingredient. Oilcakes are incorporated in animal feeds as high quality protein sources and are the most frequently used raw materials after maize. The oilseed meals used in South Africa include soybean meal (most frequently used), sunflower, cottonseed cake and meal (usage has diminished over the last three years), limited amounts of groundnut and canola meal as well as full-fat canola seed (DAFF, 2015). Since maize and soybean are mainly produced in the northern parts of South Africa or imported, they are more expensive in the Western Cape Province due to the high cost of transportation. Researchers in the Western Cape are therefore constantly looking for alternative locally produced protein and energy sources for animal nutrition (Brandt, 1998).

In South Africa there is a high demand for protein to satisfy both human and animal nutrition and this demand is likely to continue. It is expected that protein will become increasingly scarce and costly, particularly for animal nutrition. Further research is thus necessary to investigate the possibility of alternative protein sources for animal feeding (Brand *et al*., 2000a). In animal nutrition, the most frequently used protein sources are fishmeal, full-fat soybeans, soybean oilcake meal and sunflower- and cotton-oilcake meal. It is therefore of vital importance that the nutritional potential of all protein feeds that can be cultivated in South Africa are fully exploited (Brandt, 1998).

2.5.1. Soybean production and processing

Global soybean production is expected to exceed consumption in 2015/2016 for the fourth consecutive year, resulting in the continued accumulation of stock. In 2012, the area planted with soybean exceeded that of sunflower for the first time in South Africa (AFMA, 2015), and in 2015 it increased by 37% from 2014, reaching a record of 687 000 hectares. However, as a result of the drought production yields are expected to decline by 22% to 1.47 tons per hectare. Nonetheless, South Africa is expected to harvest a record soybean crop (raw material) in excess of 1 million tons.

Despite the increase in soybean production, it is likely that consumption will continue to exceed production, as the projected soybean oilcake (processed form) consumption is over 1.2 million tons, with just under 800 000 tons of soybean oilcake being produced locally. The nearly 40% shortfall will have be imported (AFMA, 2015). The importation of soybean oilcake has decreased since 2011/2012 (AFMA, 2014), and for the fourth consecutive year there was a decline in soybean imports by South Africa. During 2013/2014, 610 022 tons of soybean oilcake was imported in comparison to the 487 919 tons in 2014/2015, which represents a 20% decrease (AFMA, 2015).

2.5.2. Local soybean strategy

Around 2011/2012 the Department of Trade and Industry (DTI) and International Trade Administration Commission of South Africa (ITAC) developed a national strategy to accelerate the development of the local soybean industry. This strategy aims to increase local production at both the upstream agricultural level and downstream processors level by increasing local production from both commercial and smallholder farmers (DAFF, 2015). The establishment of the new local crushing capacity gave the industry the opportunity to channel more local soybeans to crushing for animal feed. The initial high volumes of soybean oilcake imports were replaced due to the market mechanism allowing more local soybean oilcake to be taken up by the local industry. However, South Africa remains a net importer of soybean oilcake, despite the growing local soybean oilcake production, and the domestic demand is expected to increase by 35% over the next decade (AFMA, 2015).

The South African feed industry is also faced with the following challenges in the consistent supply of local soybean oilcake meal: availability at the feed mill throughout the year, regularity of crushing, consistency of nutritional quality (especially protein content), product price *versus* international prices and the rising presence of *Salmonella* contamination at different sampling points in the production environment within the feed mills. *Salmonella*-contaminated feed ingredients are an important source for introducing *Salmonella* into the feed and food chain (DAFF, 2015). Furthermore, feed demand has increased by 42% over the past decade and a further 39% increase is expected over the next decade (AFMA, 2014).

Of all the feed components, protein sources in particular are becoming scarcer and more expensive (Brand *et al*., 2004). The trend of replacing imported soybean oilcake with local products is clear and this trend is likely to continue (AFMA, 2015). Nonetheless, the nutritional value of other locally produced plant protein sources, such as lupins and other oilseeds, need to be evaluated to see whether they can serve as economical alternatives.

2.5.3. Alternative protein sources

In 1949 the benefits of lupins as a crop that improves the soil and as an animal feed were realised. Lupins have subsequently been incorporated into crop cultivation schemes and animal feeding programmes in the winter rainfall region of the Western Cape, especially the Swartland (Van der Merwe, 1970; De Villiers, 1980). As a result, lupins (*Lupinus angustifolius*) were successfully identified as an economical alternative plant protein source that is able to be cultivated and fed to animals at a positive profit margin.

2.6. LUPINS AS AN ALTERNATIVE PROTEIN SOURCE TO SOYBEAN OILCAKE MEAL IN OSTRICH DIETS

2.6.1. Background on lupins

For over 2000 years, the seeds of various lupin species have been used in human and animal diets (Gladstones, 1970). The three species of lupins can be distinguished according to the colour of their flowers; namely *Lupinus angustifolius* (blue), *L. albus* (white) and *L. luteus* (yellow) (McDonald *et al*., 2011). The whole seed (grain) of these species are simply referred to as lupins (Petterson and Fairbrother, 1996). Between the 1950's and 1960's, researchers in Western Australia concentrated on improving the agronomic characteristics and overcoming the bitter flavour of lupins (Gladstones, 1982). More palatable varieties within the *Lupinus angustifolius* species were successfully cultivated by researches and in order to distinguish them from bitter varieties grown elsewhere, they are frequently referred to as Australian sweet lupin, which is now a major crop in Western Australia. The seed and vegetative parts of lupins are a rich source of protein and energy, and the seed can be fed as a whole or ground grain, making it an invaluable resource for both monogastric and ruminant production systems (Petterson and Fairbrother, 1996; Todorov *et al*., 1996; Edwards and van Barneveld, 1998). Currently most of the seed that is sold is used in compound feeds for intensive animal industries, but there is an increasing interest in the use of lupins in the human food industry (Petterson and Fairbrother, 1996).

In recent years, sweet lupins (*L. angustifolius* and *L. albus*) have been introduced as a legume crop in the winter rainfall region of South Africa, and it is estimated that approximately 20 000 tons of lupins are produced annually. In the Western Cape Province, lupins are an important rotation crop and approximately 15 000 hectares of lupins are cultivated annually (Brandt, 1998; Agenbag, G.A., Pers. Comm., Dept. of Agronomy, University of Stellenbosch, Private Bag X1, 7602, Matieland, South Africa, 25 Oct. 2014). According to Todorov *et al*. (1996), lupins are very productive and highly adaptable plants and can thus be cultivated under a wide variety of ecological conditions. Lupins can also be used very efficiently in crop rotational systems, as they do not have

any specific requirements as to their location in the system. However, in low-land rotations they are usually cultivated after wheat or other grains, while in high-land rotations they are planted after oats, rye or potatoes (Todorov *et al.*, 1996). In crop rotational systems, lupins can be used as a nitrogen-fixing legume, and to provide a useful break in the build-up of diseases in cereals.

Lupins are cost-competitive with multiple other protein sources, making them a widely used protein and energy source in livestock feeds (Edwards and van Barneveld, 1998). Only soybean can compete with lupins in this respect. Lupins can therefore serve as a valuable alternative to soybeans (Petterson and Fairbrother, 1996).

2.6.2. Chemical composition and nutritional value

As can be seen in Figure 2.5 and 2.6, a large part of the nutrient composition of both soybean oilcake meal and lupins is protein, with soybean oilcake meal containing 55% protein (Ewing, 1997) and lupins 33.8% (Feedipedia, 2016). Lupins also have a relatively high oil content in comparison with soybean oilcake meal, which contributes to their high energy value (Todorov *et al.*, 1996). Lupins have 12.6% more NDF than soybean oilcake meal. NDF is a measure of the fibrous plant parts that make up the structural components (lignin, hemicellulose and cellulose, but not pectin) in plant cells. The fibrous seed coat and NDF content of lupins will affect the digestibility of the meal and is often not desired (McDonald *et al.*, 2011). This is a major concern for young monogastric animals (Brand, 1996), but the ostrich has the ability to digest fibrous material. This is due to their capacity for hindgut fermentation, which distinguishes them from other monogastric herbivores (Brand and Gous, 2006). The high protein content of lupins makes them an invaluable resource for both monogastric and ruminant production systems (Edwards and van Barneveld, 1998).

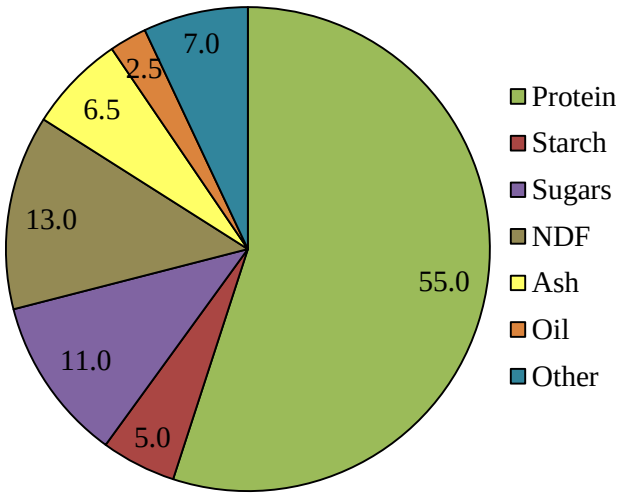

Figure 2.5. *Nutrient composition of soybean meal (%) (Ewing, 1997)*

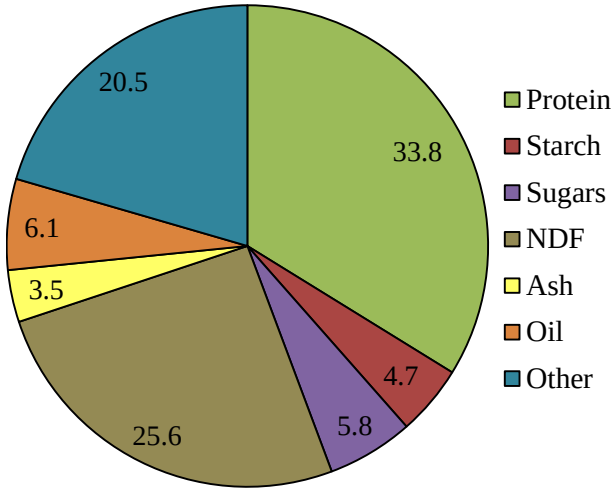

Figure 2.6. *Nutrient composition of lupins (%) (Feedipedia, 2016)*

The amino acid and mineral profiles of the three lupin species are presented in Table 2.8. Lupins contain relatively low levels of lysine and methionine and very high levels of arginine. The sulphur-containing amino acids, required by animals for a balanced diet, are insufficient in all legumes' seeds (McDonald *et al*., 2011; Edwards and van Barneveld, 1998). Diets that contain significant amounts of lupin meal therefore need to be supplemented with methionine, due to the imbalance in the amino acid profile (McDonald *et al*., 2011). Todorov *et al*. (1996) reported that supplementing methionine significantly increased the efficiency of protein utilisation in protein isolates and concentrates. However, the low methionine levels can limit wool production in sheep and milk production in highly productive dairy cattle and it should thus be supplemented (Agenbag, 2008). Furthermore the low methionine and lysine levels will influence the growth and production of monogastric animals, but these shortfalls can be supplemented by other proteins rich in these amino acids or synthetic amino acids (Edwards and van Barneveld, 1998). Lupins contain acceptable phosphorous levels, very low calcium levels and copper and zinc levels that are below the requirements of animals, but these deficiencies can be supplemented (Agenbag, 2008).

Agenbag (2008) noted that lupin seed can be successfully included in the diets of sheep, cattle, pigs, poultry and ostriches. Even the harvest residue (stems, leaves, legume pods and seeds) can serve as valuable grazing for sheep and cattle during the dry summer months when good quality grazing is limited. Lupin stubble may in fact be of a better quality than cereal stubble (Agenbag, 2008). Lupins are regarded as a highly desirable ruminant feed due to their low starch levels and high levels of fermentable carbohydrates. They serve as an excellent substrate for microbial fermentation and have a low incidence of causing acidosis (McDonald *et al.*, 2011). Diets with a high lupin content can be fed to sheep, cattle and goats with little risk of acidosis, as ruminants are able to break down non-starch polysaccharides (NSPs) without any build-up of lactic acid (Petterson and Fairbrother, 1996). The dehulling of lupins for ruminants is also unnecessary. Lupins can be fed to cattle in a ground or rolled form, while sheep utilise whole lupins very efficiently (Dixon and Hosking, 1992).

While all the chemical components of lupins are fairly digestible for ruminants, monogastric animals, such as horses, poultry, and pigs, lack the enzymes necessary to digest the carbohydrate complex in the stomach and intestines. The digestible energy generated is therefore far less in monogastric animals than in ruminants (Agenbag, 2008). However, as previously discussed, the ostrich has a relatively large digestive track, providing an ideal environment for the fermentation of fibrous material (Brand and Gous, 2006).

Table 2.8. *The chemical, mineral and amino acid composition (as fed basis) of the three species of lupin versus soybean oilcake meal (adapted from Ensminger et al. (1990) and Agenbag (2008))*

Nutrient component (g/kg)	Narrow leaf (*Lupinus angustifolius*)	Broad leaf (*Lupinus albus*)	Yellow lupins (*Lupinus luteus*)	Soybean oilcake meal (*Glycine max*)
Dry matter	916.0	914.0	917.0	890.0
Crude protein	339.0	382.0	394.0	444.0
Ash	31.0	36.0	44.0	64.0
Ether extract	58.0	91.0	50.0	15.0
Acid detergent fibre	197.0	170.2	209.0	89.0
Neutral detergent fibre	227.0	173.1	254.0	125.0
Minerals				
Calcium (g/kg)	1.9	1.6	1.7	3.5
Phosphorous (g/kg)	4.2	4.5	7.6	6.4
Magnesium (mg/kg)	0.2	0.2	0.3	2.7
Copper (mg/kg)	3.4	5.0	5.0	19.9
Zinc (mg/kg)	34.6	33.9	55.3	50.5
Iron (mg/kg)	50.0	33.7	53.7	0.02
Amino acids (g/kg)				
Lysine	14.3	15.7	20.2	28.5
Methionine	2.20	2.40	2.80	5.90
Cysteine	4.60	5.00	9.10	6.70
Arginine	36.5	46.8	44.9	32.6
Threonine	10.4	11.9	14.5	18.1
Tryptophan	3.20	3.70	3.80	6.20
Tyrosine	10.7	17.1	10.8	6.00
Histidine	7.60	6.50	10.9	11.3
Valine	11.8	13.6	14.8	23.7
Phenylalanine	11.2	12.3	16.0	22.3
Isoleucine	12.3	14.1	15.9	21.2
Leucine	20.8	23.0	27.6	34.9

Pigs and poultry require specific levels of individual amino acids, but the crude protein content is often used as a guide for the amino acid profile rather than direct amino acid analysis. Unfortunately this is not ideal as the effective use of lupins in pig and poultry diets can be affected by variation in both the crude protein content and the amino acid composition.

The complex carbohydrate profile of lupins is the main constraint on their use in monogastric diets, as it influences the net energy yield and affects the utilisation of the other nutrients in the diet (Edwards and van Barneveld, 1998). Dehulled lupins can be included in monogastric diets at higher levels. It is however essential that only alkaloid-free cultivars are included in the diets of monogastrics, due to the anti-nutritional effects of these compounds (Dixon and Hosking, 1992). Alkaloids are bitter tasting compounds, reducing the palatability of the feed (Smith, 2005).

2.6.3. Nutritional value per animal species

Sheep

During the dry season, when fresh fodder or grazing is not available, lupin meal can be used as a supplementary feed for both young animals and pregnant or lactating ewes. High-grain diets are also fed to early-weaned lambs and growing and feedlot sheep (Hodge *et al.*, 1981). Thus, high productivity and normal growth can be maintained by feeding lupin grain and meal. The birth weight of lambs and their growth rates can be increased by supplementing the diet of pregnant ewes with 400 g/day for two weeks prior to lambing (Todorov *et al*., 1996).

A higher feed intake, live weight change and wool growth are generally achieved by supplementing lupin grains rather than comparable supplements of cereal grains. Dixon and Hosking (1992) reported that this was primarily due to the protein content of lupins, which provides nitrogen for microbial protein synthesis. However, it might also be partly attributable to the bypass protein effect, higher ME content and reduced disturbance of fibre digestion, which often accompanies the fermentation of cereal starch.

Lupins are also commonly fed to grazing sheep to improve their fertility and fecundity. Feeding rams 200 g/day for eight weeks prior to mating provides sufficient energy and protein for increased sperm production, leading to an increase in their testicular size and fertility. Higher lambing percentages are attained by feeding ewes 400-500 g/day for two weeks prior to and after mating (Edwards and van Barneveld, 1998).

Dairy and beef cattle

Lupin grain and meal can be successfully fed to dairy cows to improve their productivity (Todorov *et al*., 1996), but are seldom fed as a sole supplement. According to Edwards and van Barneveld (1998), lupins should rather be fed in a mixture with cereal grains and other feedstuffs. The inclusion level of lupins in the diet depends on the protein content of the pasture or the available fodder, the lactation stage and production level and the cost of lupins relative to cereal grains and other protein meals. The use of animal protein supplements has been banned from dairy diets, due to consumers' concerns regarding *Bovine spongiform encephalopathy* (BSE), better known as mad cow disease (Jayasena and Quail, 2004). The emphasis has therefore shifted to plant protein sources, such as canola and cottonseed meal, to provide the necessary bypass protein for high-producing dairy cows (Edwards and van Barneveld, 1998).

Rumen microbial protein synthesis is sufficient for most cattle to meet their protein requirements for full growth potential, with the exception of young, fast-growing cattle in a feedlot system. During the introductory phase of feedlotting, lupins may have an economical role, because a gradual transition from lupin grain to cereal grain allows higher initial levels of feeding and subsequent growth (Edwards and van Barneveld, 1998). Productivity can also be successfully enhanced in beef steers by adding lupin grain and meal to their diet (Todorov *et al.*, 1996). Lupin grains will be most competitive when relatively cheaper than cereal grains or when the convenience and safety of feeding is highly valued (Petterson and Fairbrother, 1996).

Lupins are most valuable when they can provide a source of additional energy and protein, such as when supplementing low quality pasture or roughage. On high quality pasture or silage, the value of supplementation with lupins may be limited, as the animals will most likely not be lacking in plant protein. The following factors determine to what extent lupins are utilised in beef cattle feeding: the composition and abundance of the pasture or conserved fodder (basal feedstuff) and the price and availability of lupins in relation to alternative feedstuffs (Edwards and van Barneveld, 1998).

Pigs and poultry

Currently the preferred lupin species for pig diets is the Australian sweet lupin (Petterson and Fairbrother, 1996). Diet formulations must precisely meet the requirements of the pig and the available amino acid and energy content of a feedstuff is therefore important (Edwards and van Barneveld, 1998). The maximum inclusion levels recommended by Australian researches are as follows: 10-15% in starter diets, 20-25% in grower diets, 30-35% in finisher diets and 20% in dry and lactating sow diets (Petterson and Fairbrother, 1996). A lower intake and growth rate can be expected due to the higher fibre content of lupins than other protein sources (Brand *et al*., 1995).

Petterson and Fairbrother (1996) noted that *L. albus* has some palatability concerns and is not recommended for use in pig diets, due to poor acceptance and growth rates. Lysine and methionine supplementation is essential when a lupin-based diet is fed to pigs and poultry in order to provide a balanced ration (Petterson and Fairbrother, 1996)

Inclusion levels of up to 25% of low-alkaloid lupin-seed meal can be tolerated by broiler chickens without affecting growth unfavourably, but adequate lysine and methionine must be supplemented (Brenes *et al.*, 1993). Research has shown that a maximum inclusion level of 25-35% of either *L. angustifolius* or *L. albus* will not affect the laying performance of hens (Edwards and van Barneveld, 1998). However, the inclusion level of either *L. angustifolius* or *L. albus* in broiler chicken diets should not exceed 10%. This is to prevent the incidence of wet-sticky droppings, which may be promoted by high levels of lupin NSPs. This contributes to higher litter moisture levels, which causes respiratory stress from increased ammonia levels and may result in breast blisters which will decrease carcass quality. There is also a greater risk of coccidiosis. Litter disposal can also cause environmental issues (Edwards and van Barneveld, 1998).

Brand *et al*. (2004) determined and compared the physical characteristics, chemical composition and energy values of the different types of lupins, faba beans and narbon beans for poultry and showed that large variations occurred in the composition and energy content of grain legumes. High inclusion levels of these grain legumes, especially in broiler diets, are limited due to the high fibre levels as well as the anti-nutritional factors which may also be detrimental for the utilisation of legume grains.

2.6.4. Anti-nutritional factors of lupins

Breytenbach (2005) investigated *L. angustifolius* and found that there are sweet (low in alkaloids, < 0.1%) and bitter (alkaloid-rich, 0.1-4%) varieties within this species. To prevent any risk, an alkaloid level of less than 0.06% is deemed acceptable for use in animal feeds (McDonald *et al.*, 2011). Sweet lupin varieties with low alkaloid levels have successfully been cultivated by plant breeders in Western Australia.

Thus, with respect to anti-nutritional factors, sweet lupins are regarded as very safe and heat treatment and processing is not necessary to destroy the factors that can reduce protein digestion and availability (Allen, 1998). The prolonged intake of these varieties should be of no concern regarding toxicity for the animal (Breytenbach, 2005). While other anti-nutritional factors do occur in lupins, their risks have also been successfully reduced to manageable levels by plant breeders (Allen, 1998). Various processing procedures are also used to destroy these factors, making lupins one of the most widely used protein sources (Todorov *et al.*, 1996).

Quinolizidine alkaloids

The flowers and leaves of most lupin species have the ability to synthesise and accumulate alkaloids (Todorov *et al.*, 1996). Alkaloids are bitter compounds that have a negative effect on feed palatability for some animal species (Smith, 2005). All aerial parts and seeds of poisonous species are toxic to some extent. Nervousness, depression, difficulty in breathing, loss of muscular coordination, reluctance to move and twitching of the legs and other muscles are all signs of poisoning (Todorov *et al.*, 1996).

In sheep, symptoms include frothing at the mouth, convulsions, coma and death. When alkaloid levels are too high in pig diets, their voluntary feed intake is reduced, which results in growth depression (Allen, 1998). Lupin meal containing an alkaloid content below 0.1% can be tolerated by poultry. If levels are above 0.1%, there will be a reduction in the live weight gain of the growing chickens and the productivity of laying hens will be depressed (Todorov *et al*., 1996).

Oligosaccharides

Oligosaccharides pass unchanged through the small intestine of monogastric animals and into the lower gut, as monogastric digestive systems lack the enzyme α-galactosidase. In the lower gut, oligosaccharides are cleaved by bacterial α-galactosidase and the product is then rapidly converted to carbon dioxide, hydrogen and methane. This may result in flatulence, diarrhoea, nausea, cramps and general discomfort (Breytenbach, 2005).

Traditional debittering processes using water do not only remove the alkaloids from lupins, but also the oligosaccharides (Allen, 1998). Heat treatment also makes oligosaccharides somewhat more digestible (Breytenbach, 2005).

Non-starch polysaccharides (NSPs)

NSPs are complex compounds and their structure is not yet fully understood. Water-soluble NSPs make up approximately 5% of the lupin seed, while the insoluble NSPs contribute an additional 30%.

The water-soluble NSPs have an anti-nutritional effect because of their viscous nature, which influences the intestinal passage rate. They also alter hormonal regulation by causing differential nutrient absorption rates (Smith, 2005). The high gut viscosity, which directly impairs nutrient absorption, decreases the rate of diffusion of substrates and digestive enzymes. It also interferes with their effective interaction at the mucosal surface. The incidence of wet-sticky droppings in poultry is also related to the soluble polysaccharide content of the feed (Breytenbach, 2005).

In contrast, the insoluble NSPs have a limited effect on nutrient utilisation by monogastric animals. Insoluble NSPs have the important characteristic of being able to hold large quantities of

water but still maintain normal gut motility (Smith, 2005). Reduced viscosity of the gut content and improved nutrient absorption can be accomplished by adding enzymes that depolymerise the soluble NSPs (Breytenbach, 2005).

Saponins

Relative to other pulses (also legumes, but referring only to the dried seeds), lupins have a very low saponin concentration. Saponins are glycosides known for having a bitter taste, foaming properties and the ability to haemolyse red blood cells (Jezierny *et al*., 2010). In addition, the irritation of the mucous membranes of the mouth and gut wall causes a depressed voluntary feed intake. This leads to growth inhibition and an increased excretion of cholesterol. This is of particular concern for monogastric animals (Petterson and Fairbrother, 1996; Allen, 1998).

Tannins

Tannins bind with digestive enzymes and inhibit their activity, which causes a reduction in protein utilisation (Smith, 2005). Tannins are polyphenolic compounds that may either bind with proteins in the intestinal mucosa to affect general nutrient absorption or with vitamin B12 to make it unavailable for absorption. The reaction between tannins and the salivary proteins and glycoproteins in the mouth creates a strong bitter taste. Palatability and feed intake is thus reduced, the gastrointestinal tract experiences irritation, the energy conversion efficiency from the feed is reduced and there is an increase in nitrogen excretion. However, lupins contain a lower concentration of tannins than soybeans (Allen, 1998).

Protease/Trypsin inhibitors

The two most common protease inhibitors are trypsin and chymotrypsin inhibitors (Allen, 1998). Although the concentrations of these inhibitors are very low in lupins relative to other legumes, they can still interfere with digestive processes (Petterson and Fairbrother, 1996; Allen, 1998). The presence of trypsin inhibitors can also result in poor growth, reduced feed intake and protein digestibility, pancreatic hypertrophy and a deficiency of sulphur-containing amino acids (Allen, 1998). However, one of the primary advantages of lupins is their low content of the aforementioned biologically active non-nutritive components.

2.7. CONCLUSION AND OBJECTIVES

For more than 150 years, ostrich farming has been practiced in South Africa and despite being continuously faced with challenges, it continues to grow rapidly. A considerable amount of research has been conducted on various aspects of the industry to improve the profitability of an intensive ostrich enterprise. However, the most important cost component of an ostrich production unit is the feeding costs. In order to decrease feeding costs and ensure the cost-efficient production of slaughter ostriches, without affecting their growth performance, producers need to find alternative protein sources to soybean oilcake meal which can be cultivated locally at lower costs. Nutritionally, lupin seed (grain) could offer some advantages as an alternative to soybeans. Therefore, this review aimed to gather information contributing to ostrich nutrition and apply these findings to the planned research on the use of lupins in ostrich diets in the best interest of the industry.

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CHAPTER 3

THE EFFECT OF VARYING SWEET LUPIN DIETARY INCLUSION LEVELS ON THE PRODUCTION AND SLAUGHTER TRAITS OF OSTRICHES (*Struthio camelus* var. *domesticus*)

Abstract

1. Nutrition contributes approximately 75% of the total input costs of an intensive ostrich production unit. An increase in the price of traditional protein sources necessitates finding cheaper alternatives. Sweet lupins were identified as a possible alternative and therefore the effect of feeding various sweet lupin inclusion levels was evaluated throughout the different feeding phases.

2. One hundred and forty ostrich chicks were randomly divided into five dietary treatments with three replications each. The chicks all received a standard commercial pre-starter ostrich diet. Five iso-nutritional diets were formulated for each feeding phase according to specifications set out for each of the different feeding phases using Mixit2+ software. For each feeding phase the diets contained either soybean oilcake meal (control diet, 0LD) or sweet lupins (alternative protein source). The 100 lupin diet (LD) was formulated to include the maximum amount of sweet lupins according to the specifications for the specific species and the feeding phase and therefore differs between the three feeding phases. Soybean oilcake meal was thus gradually replaced by sweet lupins in the following ratios: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) and 0:100 (100LD). Feed and water were supplied *ad libitum*. Biweekly feed intake and live weight were measured until slaughter at *ca*. 11 months of age.

3. The initial average body weight of the birds was 13.8 ± 0.55 kg. No differences were found between the treatment diets for the live weight, dry matter intake (DMI), average daily gain (ADG) or feed conversion ratio (FCR) at the end of each feeding phase. However, the birds on the 75LD tended to have the highest end weight (P = 0.07) and ADG (P = 0.09) and those on the 100LD the lowest at the end of the starter phase.

4. No differences were found for the slaughter weight, dressing percentage and big drum muscle weight of the birds. Birds on the 50LD and 75LD tended (P = 0.08) to have the heaviest cold carcasses which differed from the 100LD birds, which had the lightest. Birds fed the 50 h had (P = 0.05) heavier thigh weights than those on the other diets.

5. The results of this study indicate that soybean oilcake meal can be replaced in the diets of slaughter ostriches with sweet lupins up to 15% (75LD) in starter diets and 30% (100LD) in grower and finisher diets without any significant effect on any of the production and slaughter traits.

3.1. INTRODUCTION

The ostrich industry is still relatively new, as ostriches have not been domesticated for as long as traditional livestock species (Brand and Olivier, 2011; Cloete *et al*., 2012). Any increase in reliable scientific information will enhance the scope of opportunities and the profitability of the industry. However, for more than a century now, the South African ostrich industry has been growing and intensifying (Brand *et al*., 2002) and this trend is likely to continue since there is still a lot to discover regarding these species.

In April 2011 the ostrich industry was severely impacted by the highly pathogenic *Avian influenza* (HPAI) H5N2 outbreak which led to the European Union (EU) banning the import of fresh meat. This resulted in a drastic decrease in the gross value of ostrich meat. Since then the industry has recovered to some extent due to increases in the value of ostrich leather, the development of pre-cooked (*sous vide*) meat products that were successfully exported and an increase in the local consumption of ostrich meat (DAFF, 2014; Booysen, 2015). In August 2015 the European Commission announced the lifting of the four-year ban on the import of fresh ostrich meat into the EU. The industry contributes over R1 billion to the economy annually and currently employs over 50 000 people. The reestablishment of the export market for fresh meat will allow further growth in the South African ostrich industry (Booysen, 2015).

On the farm level, high running costs remain a problem, with nutrition contributing *ca*. 75% of the total input costs of an intensive ostrich production unit (Brand and Jordaan, 2011). A large part of these costs are represented by the protein source, with soybean oilcake meal being the primary protein source in animal feeds (Dalle Zotte *et al*., 2013). The increase in the demand for traditional protein sources and thus the price makes it necessary for producers to find cheaper alternatives to ensure the cost efficient production of slaughter ostriches while also not affecting their growth performance. The gastrointestinal tract of the ostrich develops from a typical bird neonate to that of a hindgut fermenter in a space of 70 – 80 days (Brand and Gous, 2006). Slaughter ostriches therefore generally receive the following diets: pre-starter, starter, grower, finisher and maintenance for the different feeding phases when reared intensively.

Information on the nutritive value of alternative protein sources in ostrich diets is limited (Brand *et al*., 2000a). This necessitates the quantification of the nutrient composition of raw materials in order to allow the formulation of diets that more closely fit the needs of the bird and to ensure the continued economic viability of the industry. Thus, to improve the feeding regimes of ostriches the inclusion of alternative protein sources such as lupins (*Lupinus angustifolius*) need to be investigated. Three species of lupin are distinguished according to the colour of the flowers, namely *L. angustifolius* (blue), *L. albus* (white) and *L. luteus* (yellow) (McDonald *et al*., 2011). There are also sweet (low in alkaloids, < 0.1%) and bitter (alkaloid-rich, 0.1 - 4%) varieties within

these species (Breytenbach, 2005). Sweet lupin varieties with low alkaloid contents have successfully been cultivated by plant breeders, therefore the use of these varieties in diets with prolonged intakes should be of little concern regarding toxicity for the animal. However, lupins have not yet been fed to ostriches as part of a balanced feedlot diet. Therefore the inherent characteristics of lupins, such as the anti-nutritional factors (e.g. alkaloids and non-starch polysaccharides) and high fibre levels that could influence the production and slaughter traits of the birds need to be evaluated to determine the optimum lupin inclusion level per feeding phase.

This study therefore aimed to determine whether and to what extent soybean oilcake meal can be replaced by locally produced sweet lupin seed in the diets of ostriches (*Struthio camelus* var. *domesticus*).

3.2. MATERIALS AND METHODS

3.2.1. Experimental location

Ethical approval (R14/108) for this trial was obtained from the Western Cape Department of Agriculture. The trial was carried out from November 2014 until September 2015 at the Kromme Rhee Research Farm near Stellenbosch in the Western Cape Province of South Africa. The farm is situated at longitude 18°50' E and latitude 33°51' S with an altitude of 177 m above sea level.

3.2.2. Experimental design and animal management

The ostrich chicks used in this trial were obtained from the resource flock at the Oudtshoorn Research Farm in the Little Klein Karoo region of South Africa (situated at longitude 22°15' E and latitude 33°37' S at an altitude of 300 m above sea level) and transported after hatching to the experimental site. On arrival, the chicks were randomly divided into their five dietary treatment groups with three replications each, resulting in 15 groups of chicks with 15 – 17 chicks per group. The chicks were initially reared indoors in 15 pens of 2.85 m x 2.05 m per pen with free access to outdoor runs (14.60 m x 1.80 m) during the day. In the late afternoon the indoor pens were closed to protect them against the elements. At nine weeks of age the chicks were moved into outdoor paddocks of approximately 25 m x 20 m to provide sufficient space to avoid unnecessary skin damage and leg abnormalities. The outdoor paddocks were equipped with adequate shelter for the ostriches and feed troughs (67 cm x 60 cm x 78 cm). The shelter prevented the feed from getting wet from dew or rainfall. Mortalities were recorded and dead birds were send for post-mortem investigation throughout the experimental period to ensure that the dietary treatments were not responsible for these deaths. Most of the mortalities (37.3%) occurred during the pre-starter phase and were mainly due to fractured legs, tibio-tarsal deformities, haemorrhagic enteritis, yolk sac

infection, torsion of the intestine and gastric stasis and dilatation; none were linked specifically to diet.

3.2.3. Dietary treatment

During the lifetime of a slaughter ostrich under intensive conditions, it receives the following diets: pre-starter, starter, grower and finisher (Brand and Gous, 2006; Brand and Olivier, 2011). Although the chicks were divided into their experimental groups on arrival at the farm, they all received the same standard commercial pre-starter ostrich diet (Table 3.1).

The trial period started at the onset of the starter phase, at which point the 141 surviving ostrich chicks were 83 days old. Five iso-nutritional diets were formulated using Mixit2+ software (Agricultural Software Consultants Inc., San Diego, USA) for each feeding phase (i.e. starter, grower and finisher) according to specifications predicted by a model developed by Gous and Brand (2008). For example a starter phase chick does not have the ability to digest the same amount of fibre that a grower and finisher bird can utilise. Their ability to digest crude fibre efficiently only develops after approximately three months of age (large intestine is not fully developed yet) (Angel, 1996). This gives rise to the need for different feeding phases at specific growth stages when ostriches are reared intensively. Within each feeding phase these diets contained either soybean oilcake meal (control diet; 0LD) or sweet lupins (alternative protein source; diets 25LD, 50LD, 75LD and 100LD) as the primary protein source.

Table 3.2 contains the raw material composition of the sweet lupin and soybean oilcake meal used in the diet formulations of this study. These values were determined by grounding the raw material samples using a RetschTM ZM200 sample mill (Haan, Germany) with a 1.5 mm screen to create a meal with a consistent particle size. Thereafter the raw materials were analysed using the methods of the Association of Official Analytical Chemists (AOAC, 2002) for dry matter (DM) (method 934.01), ash (method 942.05), crude protein (CP) (method 976.05), crude fibre (CF) (method 962.09), ether extract (EE) (method 920.39), acid detergent fibre (ADF) (Goering and van Soest, 1970), neutral detergent fibre (NDF) (Robertson and van Soest, 1981). The calcium (Ca) and phosphorous (P) values were analysed using method 6.1.1 (Dry Ashing) of the Agri Laboratory Association of Southern Africa guidelines (ALASA) (ALASA, 1998).

The 100 lupin diet (LD) was formulated to include the maximum amount of sweet lupins according to the specifications for the specific species and the feeding phase. The maximum amount of lupins included in the 100LD therefore differs between the three feeding phases. The remaining three diets are formulated by mixing the diets to determine the gradual increase in lupins in the diets from 0LD up to 100LD (Table 3.3).

Table 3.1. *The formulation and nutritional composition (as fed basis) of the pre-starter diet fed to all the experimental groups (kg/ton) from 0 – 83 days of age*

Raw materials (kg)	Dietary composition
Maize meal	484.4
Soybean oilcake meal	185.0
Wheat bran	150.5
Fishmeal	75.0
Bentonite	25.0
Plant oil	40.0
Monocalcium phosphate	1.90
Limestone	21.3
Salt	10.0
Synthetic lysine	1.95
Vitamin and vitamin premix*	5.00
Nutrient composition	
Metabolisable energy (MJ/kg feed)	14.5
Crude fibre (g/kg)	71.5
Ether extract (g/kg)	53.6
Crude protein (g/kg)	184.7
Lysine (g/kg)	8.80
Methionine (g/kg)	1.13
Threonine (g/kg)	4.09
Arginine (g/kg)	8.34
Ash (g/kg)	80.9
Calcium (g/kg)	12.6
Phosphorous (g/kg)	8.10
Dry matter (g/kg)	905.7

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for starter ostriches

Table 3.2. *Raw material composition of the sweet lupin and soybean oilcake meal used in the diet formulations of this study*

Nutrient component (g/kg)	Sweet lupins	Soybean oilcake meal
Dry matter	902.5	910.8
Ash	29.5	62.5
Crude protein	309.4	463.1
Crude fibre	154.0	32.0
Ether extract	48.9	10.3
Neutral detergent fibre	244.8	81.9
Acid detergent fibre	196.9	44.3
Calcium	2.60	2.90
Phosphorous	4.90	8.30

Table 3.3. *Percentage dietary sweet lupin and soybean oilcake meal inclusion per treatment diet*

Phase	Dietary sweet lupin and soybean oilcake meal inclusion (%) per diet				
	0LD	25LD	50LD	75LD	100LD
Starter					
Sweet lupins	0	5	10	15	20
Soybean oilcake meal	18	14.7	15.5	8.5	5
Grower					
Sweet lupins	0	7.7	15.3	22.7	30
Soybean oilcake meal	14.9	11.2	7.5	3.7	0
Finisher					
Sweet lupins	0	6.7	13.3	19.9	26.6
Soybean oilcake meal	8.3	6.2	4.1	2.1	0

Soybean oilcake meal was thus gradually replaced by sweet lupins (*Lupinus angustifolius*, Eureka cultivar) in the following ratios: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) and 0:100 (100LD) to make up the five dietary treatments for each feeding phase (Tables 3.4 – 3.6). The ostriches in this trial were fed their respective diets *ad libitum* throughout the different feeding phases and according to standard practices. They also had free access to clean, fresh water throughout the trial period. In the results and discussion sections diets 1 to 5 will be referred to as 0LD, 25LD, 50LD, 75LD and 100LD for ease of identification. LD represents lupin diet and the

number indicates the ratio of the lupin inclusion level to the amount of soybean oilcake meal in the diet. The results obtained for each of the three feeding phases were statistically analysed per phase, because the lupin inclusion levels in the diets differed between the phases.

3.2.4. Alkaloid content of the feed

The total alkaloid content of the finely ground pooled feed samples of the diets containing sweet lupins (*Lupinus angustifolius*, Eureka cultivar) were determined as described by Boschin *et al*. (2008), with minor modifications to the method. The sample preparation method was modified to extract the total alkaloid content by directly using a 50:50 methylene dichloride:methanol (MDC:MeOH) mixture. The total alkaloid content was then analysed using GC-MS analysis with a 30 m x 0.25 mm, internal diameter 0.25 μm, AT-Wax capillary column. The following temperature settings were used: 150°C for 5 minutes, increased by 5°C per minute up to 300°C, maintained for 15 minutes at 300°C. Analysis was performed in split mode with a split ratio of 1:25. The injection volume was 1 μL, injection temperature 250°C, interface temperature 300°C and the acquisition was from *m*/*z* 50 to 450. The source operated in EI mode at eV. The total alkaloid content was determined using Mass library (Agilent) and the detection limit used to quantify the total alkaloids was 100 ng/ml. However, no alkaloids were found in the respective feed samples at this detection limit.

The sweet lupin cultivar used in this study was the same cultivar (*Lupinus angustifolius*) used by Brand and Smith (2016). The spectrophotometry method described by Von Baer *et al*. (1978) was used to determine the total alkaloid content of the sweet lupin cultivar used by Brand and Smith (2016). This method is a quantitative determination of total alkaloids with bromocresol purple at 405 nm. The total alkaloid content (49.1 mg/kg) of the sweet lupin cultivar found in the study by Brand and Smith (2016) was therefore used to calculate the estimated amount of total alkaloids of the five dietary treatments of the current study (Tables 3.4 – 3.6).

Table 3.4. *The formulation and nutritional composition (as fed basis, kg/ton) of five treatment diets containing sweet lupins at different inclusion levels fed to starter phase slaughter ostriches from 83 – 167 days of age*

Raw materials (kg)	Diet (SOM replacement) and sweet lupin inclusion level (%)				
	0LD (0%)	25LD (5%)	50LD (10%)	75LD (15%)	100LD (20%)
Maize meal	568.6	550.5	532.5	514.4	496.3
Soybean oilcake meal (SOM)	180.0	147.5	115.0	82.5	50.0
Sweet lupins	0.00	50.0	100.0	150.0	200.0
Lucerne meal	140.0	140.0	140.0	140.0	140.0
Plant oil	25.0	25.0	25.0	25.0	25.0
Molasses powder	25.0	25.0	25.0	25.0	25.0
Monocalcium phosphate	22.5	22.4	22.3	22.1	22.0
Limestone	23.0	23.5	24.0	24.5	25.0
Salt	10.0	10.0	10.0	10.0	10.0
Synthetic lysine	2.40	2.60	2.80	3.00	3.20
Synthetic methionine	0.00	0.00	0.00	0.00	0.00
Mineral and vitamin premix*	3.50	3.50	3.50	3.50	3.50
Nutrient composition					
Metabolisable energy (MJ/kg feed)	13.5	13.5	13.6	13.6	13.6
Crude fibre (g/kg)	58.4	60.7	63.1	65.4	67.7
Ether extract (g/kg)	52.1	56.0	60.0	63.9	67.8
Crude protein (g/kg)	155.9	156.4	156.93	157.5	158.0
Lysine (g/kg)	9.30	9.30	9.30	9.30	9.30
TSA (g/kg)**	5.05	4.96	4.88	4.79	4.70
Threonine (g/kg)	5.79	5.81	5.82	5.84	5.85
Tryptophan (g/kg)	2.01	1.90	1.78	1.67	1.55
Arginine (g/kg)	8.90	9.62	10.3	11.1	11.8
Ash (g/kg)	20.9	20.7	20.6	20.5	20.4
Calcium (g/kg)	15.0	15.0	15.0	15.0	15.0
Phosphorous (g/kg)	7.88	7.72	7.57	7.41	7.25
Dry matter (g/kg)	899.8	905.2	910.5	915.8	921.1
Total alkaloid content (ppm)	0.00	2.45	4.91	7.37	9.82

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for starter ostriches

**Total sulphur containing amino acids

Table 3.5. *The formulation and nutritional composition (as fed basis, kg/ton) of five treatment diets containing sweet lupins at different inclusion levels fed to grower phase slaughter ostriches from 167 – 251 days of age*

Raw materials (kg)	Diet (SOM replacement) and sweet lupin inclusion level (%)				
	0LD (0%)	25LD (7.5%)	50LD (15%)	75LD (22.5%)	100LD (30%)
Maize meal	590.6	544.9	499.2	453.5	407.8
Soybean oilcake meal (SOM)	149.3	112.0	74.7	37.3	0.00
Sweet lupins	0.00	76.5	152.9	226.5	300.0
Lucerne meal	186.4	193.5	200.5	210.6	220.7
Molasses powder	25.0	25.0	25.0	25.0	25.0
Monocalcium phosphate	17.9	17.2	16.6	15.9	15.2
Limestone	14.5	14.8	15.0	15.3	15.5
Salt	10.0	10.0	10.0	10.0	10.0
Synthetic lysine	0.87	0.76	0.65	0.53	0.42
Synthetic methionine	0.41	0.43	0.45	0.46	0.48
Mineral and vitamin premix*	5.00	5.00	5.00	5.00	5.00
Nutrient composition					
Metabolisable energy (MJ/kg feed)	12.8	12.8	12.8	12.8	12.8
Crude fibre (g/kg)	68.7	73.8	78.8	83.8	88.9
Ether extract (g/kg)	28.2	33.9	39.5	45.2	50.8
Crude protein (g/kg)	150.5	156.0	161.5	167.0	172.4
Lysine (g/kg)	7.67	7.67	7.67	7.67	7.67
TSA (g/kg)**	5.27	5.27	5.27	5.27	5.27
Threonine (g/kg)	5.60	5.83	6.06	6.29	6.52
Tryptophan (g/kg)	0.10	0.46	0.83	1.20	1.57
Arginine (g/kg)	8.29	9.79	11.3	12.8	14.3
Ash (g/kg)	25.8	25.8	25.7	25.7	25.6
Calcium (g/kg)	12.0	12.0	12.0	12.0	12.0
Phosphorous (g/kg)	6.89	6.66	6.44	6.21	5.98
Dry matter (g/kg)	896.4	904.7	913.0	921.3	929.6
Total alkaloid content (ppm)	0.00	3.68	7.37	11.0	14.7

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for grower ostriches

**Total sulphur containing amino acids

Table 3.6. *The formulation and nutritional composition (as fed basis, kg/ton) of five treatment diets containing sweet lupins at different inclusion levels fed to finisher phase slaughter ostriches from 251 – 314 days of age*

Raw materials (kg)	Diet (SOM replacement) and sweet lupin inclusion level (%)				
	0LD (0%)	25LD (6.65%)	50LD (13.29%)	75LD (19.94%)	100LD (26.58%)
Maize meal	290.9	292.4	293.9	295.5	297.0
Oat hulls	237.7	267.4	297.2	326.9	356.7
Wheat bran	193.9	145.4	97.0	48.5	0.00
Soybean oilcake meal (SOM)	82.7	62.0	41.3	20.7	0.00
Sweet lupins	0.00	66.5	132.9	199.4	265.8
Lucerne meal	126.0	94.5	63.0	31.5	0.00
Molasses powder	24.2	24.4	24.5	24.6	24.7
Monocalcium phosphate	12.2	12.1	11.9	11.8	11.7
Limestone	17.9	20.5	23.1	25.8	28.4
Salt	9.70	9.75	9.80	9.85	9.90
Synthetic methionine	0.00	0.25	0.50	0.74	0.99
Mineral and vitamin premix*	4.85	4.88	4.90	4.93	4.95
Nutrient composition					
Metabolisable energy (MJ/kg feed)	10.7	10.8	10.8	10.9	11.0
Crude fibre (g/kg)	135.4	135.6	135.8	136.03	136.2
Ether extract (g/kg)	23.6	28.3	33.1	37.8	42.6
Crude protein (g/kg)	126.0	127.4	128.8	130.2	131.6
Lysine (g/kg)	5.50	5.50	5.50	5.50	5.50
TSA (g/kg)**	4.04	4.04	4.04	4.04	4.04
Threonine (g/kg)	4.46	4.54	4.62	4.69	4.77
Tryptophan (g/kg)	1.72	1.59	1.45	1.32	1.18
Arginine (g/kg)	6.98	8.15	9.33	10.5	11.7
Ash (g/kg)	43.2	38.8	34.5	30.1	25.7
Calcium (g/kg)	12.0	12.5	13.0	13.5	14.0
Phosphorous (g/kg)	6.81	6.29	5.78	5.26	4.74
Dry matter (g/kg)	901.6	909.2	916.7	924.3	931.9
Total alkaloid content (ppm)	0.00	3.27	6.53	9.79	13.1

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for grower ostriches

**Total sulphur containing amino acids

3.2.5. Experimental procedure

The biweekly feed intake per paddock was measured by weighing back the refusals for the two weeks and subtracting this value from the quantity of feed offered during that period. The weights of the birds were also recorded every second week on the same day for the duration of the trial for the determination of the average daily gain (ADG). When inclement weather conditions made measurements impractical, they were postponed until weather conditions improved. The feed conversion ratios (FCR) per treatment diet were expressed as DMI per kg gain per feeding phase. The birds received their compulsory inoculation against Newcastle disease at approximately five months of age and their feathers were not clipped or plucked as they were still blood feathers at six months of age.

The birds were slaughtered at approximately 11 months of age at a registered abattoir, Mosstrich (situated at longitude 22°00' E and latitude 34°09' S) in Mossel Bay. They were moved to quarantine paddocks (tick-free environment, rodent control, no vegetation and a 3 m cleared area around each paddock) 14 days prior to slaughter as required by the European Union (EU) meat quality standards highlighted in the report by the Department of Agriculture, Forestry and Fisheries (Maja, 2012; DAFF, 2014). Transport to the abattoir was performed by private contractors and on arrival the ostriches were offloaded at the specifically designed lairage area. The birds received fresh clean water during lairage and were kept in lairage for about 16 hours before slaughter commenced early the following morning.

3.2.6. Slaughtering procedures

The slaughter weight was determined by weighing each bird on an electronic walk-on scale before they entered the stunning area. The slaughtering procedures used by the abattoir staff were similar to those described by Hoffman (2012). After exsanguination, the feathers of each bird were plucked and placed in individually marked bags. After skinning, the individual skins were marked and transported to the premises of Southern Cape Ostrich Tanning (SCOT), a member of the Mosstrich Group and located in the same industrial complex as Mosstrich, for further processing. Following evisceration, the abdominal fat, better known as the fat pad, was removed, collected and weighed for each bird to determine whether the increasing sweet lupin inclusion levels of the five dietary treatments had any effect on the yield.

The carcasses, consisting of the neck, wings, chest and thighs, were washed and moved into the cold (0 – 2°C) storage facilities. The carcasses were left in the cold storage facilities for 18 hours and, before deboning commenced the next morning, the cold carcass weights were recorded. These weights were used to calculate the dressing percentage. The right thigh weight of each individual ostrich was also recorded and used to calculate the contribution of the thighs to the

carcass. It was assumed that the right and left thigh weighed the same, therefore the weight of the right thigh was multiplied by two and divided by the weight of the cold carcass to calculate the percentage contribution. The weight of the big drum muscle (*Muscularis gastrocnemius*) of the right thigh of each ostrich was also recorded and divided by the weight of the right thigh to give an indication of its contribution to the total thigh weight.

3.2.7. Statistical analysis

All statistical analysis was done using the GLM (General linear model) procedure of SAS statistical software (Version 9.2, SAS Institute Inc., Cary, NC, USA). An experimental unit consisted of a group of ostriches in a paddock fed a specific diet and the paddocks were thus regarded as random replicates of the treatment diets. The Shapiro-Wilk test was performed on all data to test for deviation from normality (Shapiro and Wilk, 1965). Fisher's protected least significant differences were calculated at the 5% level to compare treatment means (Ott, 1998). A probability level of 5% was considered significant for all significance tests.

To illustrate and describe the growth patterns over the entire trial period for each treatment diet Gompertz growth models were fitted to the data and the slopes compared to assess any differences in weight gain using analysis of variance (ANOVA):

$$\text{Bird Weight (BW)} = a*\exp(-\exp(-b*(\text{Age}-c)))$$

where a = mature weight (kg), b = rate of maturing parameter and c = age of maximum growth (days) (Brand and Olivier, 2011). As the trial only commenced at the starter phase (± 80 days of age) and the ostriches were slaughtered before a mature weight equilibrium was reached, estimated Gompertz regression parameters were not of practical interpretation value and were thus not compared statistically.

For the ANOVA and regression models on live weight and DMI, fitted over the whole trial period, the weights of birds at the start of the trial were used as a covariate. To determine the effect of the treatment diet within each phase, the end weight of the previous phase was used as a covariate in the ANOVA and regression models, i.e. initial weight, end weight and DMI within each phase were adjusted for weight differences at the beginning of that phase.

DMI per bird per day per paddock was calculated for the entire growth period, as well as per phase, as the total feed intake per paddock for a given period divided by the "bird x days". The number of ostriches per paddock did not necessarily stay stable over a given period as a result of mortalities. To account for the actual number of ostriches that consumed the feed during a given period bird x days were calculated per paddock as the sum of the product of the number of birds and days over observation times. The ADG per paddock was determined from the slope of the linear regression of growth on age (in days) within each phase, as well as for the entire period. The FCR

was calculated by dividing the DMI by the ADG. Completely random ANOVA's were conducted on the production traits, ADG and FCR.

Supplementary to the ANOVA's the treatment diet sum of squares was split up using four single degree of freedom polynomial contrasts, to determine which polynomial trend (linear, quadratic, cubic and quartic) best fitted the data over protein levels in the diet. Production traits were further analysed by fitting the relevant regression functions to the data to describe the observed trends. A probability level of 5% was considered significant and these polynomials will be reported further. Where none of the polynomials were significant, the linear polynomial was used to indicate that there was no trend.

ANOVA's were also done to analyse the slaughter traits according to the experimental design where each treatment diet was completely randomly repeated in three paddocks.

3.3. RESULTS

Gompertz growth curves for weight change in ostriches on each diet over the experimental period are presented in Figure 3.1. The Gompertz equation fits the data well within the boundaries of the data (starter, grower and finisher phases) and reveals the sigmoidal growth curve for all the diets. The estimated values for the growth parameters of the Gompertz model for the different diets are presented in Table 3.7 and the production trait means for the different treatments are presented in Table 3.8.

Table 3.7. *Growth parameters of slaughter ostriches fed different diets as predicted by the Gompertz growth curve*

Diet	Gompertz model parameters			R^2 (%)	*P**
	a[1]	b[2]	c[3]		
0LD	106.7	0.0100	153.7	80.86	< 0.0001
25LD	115.0	0.0096	161.1	81.12	< 0.0001
50LD	118.3	0.0100	158.0	84.17	< 0.0001
75LD	110.3	0.0113	150.3	86.29	< 0.0001
100LD	123.0	0.0083	174.6	85.49	< 0.0001

[1]Mature weight (kg)

[2]Rate of maturing parameter

[3]Age of maximum growth (days)

**P* value indicates the significance of the correlation

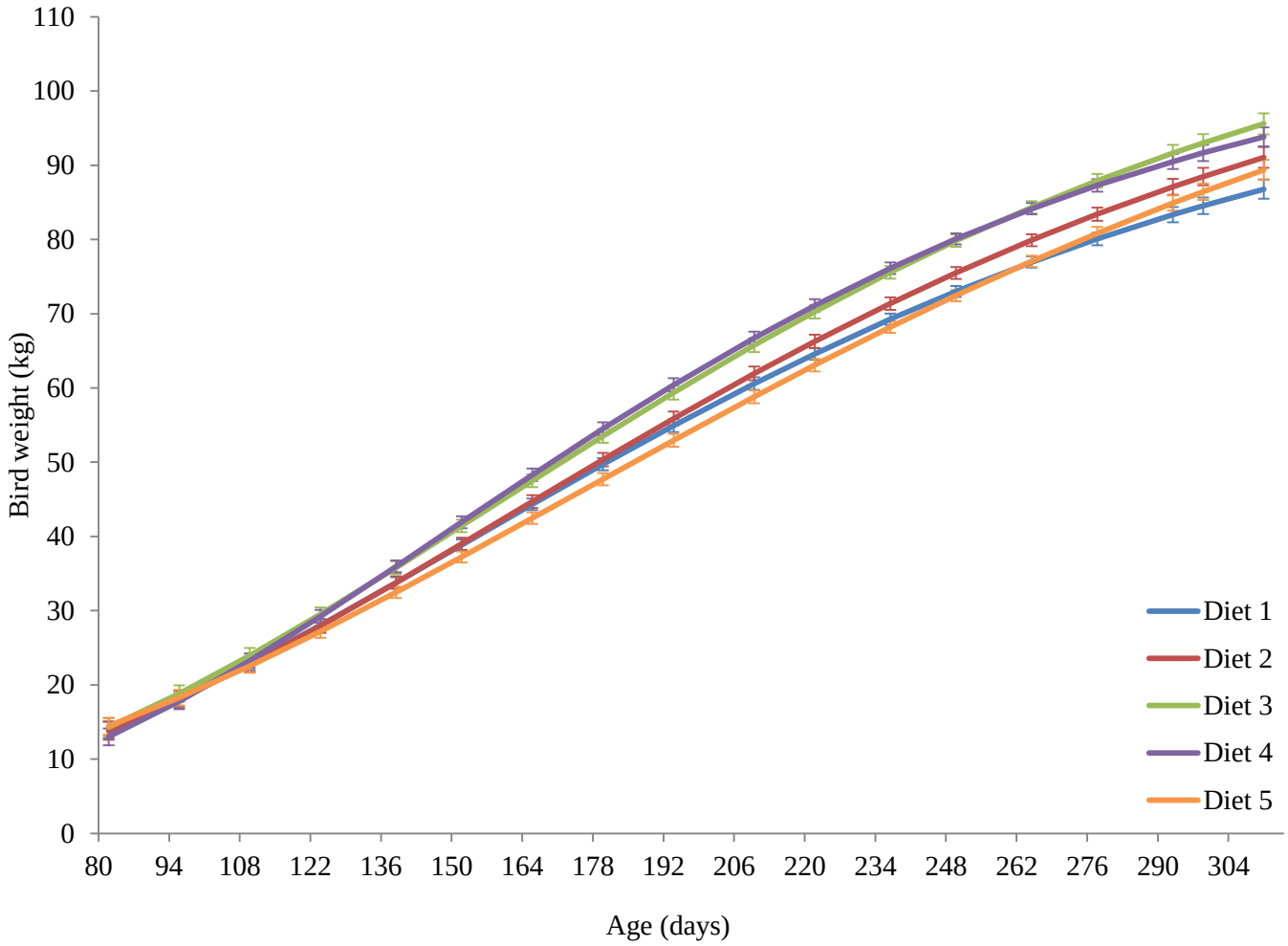

Figure 3.1. *Gompertz growth curves fitted to the mean body weights of the birds in all the paddocks per treatment diet over age (starter, grower and finisher phases)*

Table 3.8. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin inclusion levels on the production traits of slaughter ostriches*

Trait	Phase	Diet					*P*
		0LD	25LD	50LD	75LD	100LD	
Initial Weight (kg)	All Phases	13.1 ± 1.01	14.5 ± 1.01	14.5 ± 1.01	11.9 ± 1.01	14.9 ± 1.01	0.28
	Starter	13.1 ± 1.01	14.5 ± 1.01	14.5 ± 1.01	11.9 ± 1.01	14.9 ± 1.01	0.28
	Grower	40.7 ± 2.35	39.6 ± 2.35	42.2 ± 2.35	42.1 ± 2.35	37.8 ± 2.35	0.66
	Finisher	68.2 ± 3.15	72.0 ± 3.15	77.2 ± 3.15	74.5 ± 3.15	70.9 ± 3.15	0.37
End Weight (kg)	All Phases	87.5 ± 2.99	92.3 ± 3.0	94.6 ± 3.00	95.2 ± 3.39	88.9 ± 3.10	0.34
	Starter	41.9^{ab} ± 1.69	38.5^{bc} ± 1.69	41.0^{abc} ±1.69	45.3^{a} ± 1.91	35.9^{c} ± 1.75	0.07
	Grower	68.0 ± 2.17	72.9 ± 2.18	75.5 ± 2.23	72.9 ± 2.22	73.6 ± 2.30	0.25
	Finisher	89.8 ± 2.34	93.2 ± 2.15	92.1 ± 2.37	92.3 ± 2.18	90.9 ± 2.17	0.85
Dry Matter Intake (g/bird/day)	All Phases	1874.1 ± 193.4	1907.4 ± 193.8	2115.4 ± 194.0	2227.7 ± 219.0	1978.1 ± 200.3	0.70
	Starter	1104.3 ± 120.6	1020.9 ± 120.8	1233.9 ± 121.0	1317.1 ± 136.5	1095.1 ± 124.9	0.52
	Grower	1938.4 ± 271.6	2019.8 ± 273.3	2174.5 ± 278.9	2076.6 ± 277.7	2326.7 ± 288.6	0.88
	Finisher	2580.4 ± 257.1	2871.2 ± 236.0	2968.4 ± 260.2	2761.0 ± 240.1	2894.6 ± 239.0	0.56
Average Daily Gain (g/bird/day)	All Phases	324.2 ± 13.8	351.4 ± 13.8	368.7 ± 13.8	368.3 ± 13.8	344.6 ± 13.8	0.21
	Starter	405.3^{ab} ± 24.8	362.6^{ab} ± 24.8	390.5^{ab} ± 24.8	437.3^{a} ± 24.8	328.7^{b} ± 24.8	0.09
	Grower	324.4 ± 28.8	359.6 ± 28.8	402.0 ± 28.8	369.7 ± 28.8	385.2 ± 28.8	0.44
	Finisher	279.6 ± 33.5	292.0 ± 33.5	262.0 ± 33.5	283.4 ± 33.5	283.2 ± 33.5	0.98
Feed Conversion Ratio (kg feed/kg weight gain)	All Phases	5.62 ± 0.15	5.61 ± 0.15	5.91 ± 0.15	5.58 ± 0.15	6.01 ± 0.15	0.96
	Starter	2.65 ± 0.40	2.91 ± 0.40	3.33 ± 0.40	2.75 ± 0.40	3.57 ± 0.40	0.47
	Grower	6.09 ± 0.64	5.59 ± 0.64	5.57 ± 0.64	5.76 ± 0.64	5.81 ± 0.64	0.98
	Finisher	9.25 ± 1.86	9.81 ± 1.86	12.7 ± 1.86	10.1 ± 1.86	10.6 ± 1.86	0.73

[a,b,c] Row means with different superscripts differ significantly ($P < 0.10$); only indicated when means differed significantly

The average initial body weight of the birds was 13.78 ± 0.55 kg (LSM ± SE) and no differences were observed between treatment groups in the initial weight of the birds for each feeding phase (Table 3.8). The mean weight of the birds at the end of the entire experimental period (all phases) was 91.7 ± 1.52 kg and this did not differ (P = 0.34) between the respective dietary treatments. Furthermore, no differences were found between the treatment diets for the weights of the birds at the end of each feeding phase. However, there was a tendency (P = 0.07) for the live weights at the end of the starter phase to differ between the treatments. The mean end weight of the birds on the 75LD was 26.2% heavier than that of the birds fed the 100LD, who were the lightest (35.9 ± 1.75 kg) at the end of the starter phase. Regarding the production traits for the starter, grower, finisher and overall growth phases (Table 3.8), no differences were found between the treatment diets for the DMI, ADG or FCR. The only exception to this was the tendency (P = 0.09) observed in the ADG during the starter phase. The live weight of the birds on the 75LD increased by 33.0% and in comparison to the birds on the 100LD, which had the lowest gain during this period. Mean DMI and ADG of 2020.5 ± 61.9 g/bird/day and 351.5 ± 8.3 g/bird/day respectively were observed over all the treatments, with a mean FCR of 5.75 ± 0.09 kg feed/kg weight gain found for the whole trial period (all phases).

Production traits were further analysed using various regression models to describe trends resulting from changes in the sweet lupin inclusion levels in the diets over all phases and within each phase of the trial (starter, grower and finisher) separately. A probability level of 5% was considered significant and the regression models corresponding to the most significant, polynomial contrasts (Table 3.9) are presented in Table 3.10. Where none of the polynomials were significant, the linear polynomial was used to indicate that there was no trend and is also presented in Table 3.10. The only significant trends (P < 0.05) in the regression models were for end weights in the starter, grower and finisher phases. A cubic function (R^2 = 62.15% and P = 0.03) best fitted the end weights of the starter phase (Figure 3.2), whereas for the grower (R^2 = 59.44% and P = 0.005; Figure 3.3) and finisher (R^2 = 62.82% and P = 0.003; Figure 3.4) phases, a linear function fitted the end weight over diet relationship.

Regression analysis of the remaining three production traits (DMI, ADG and FCR) per phase and over the entire experimental period revealed no significant trends. However, the regression model for the ADG over the whole growth period and during the starter phase was significant (P < 0.05). Regression analysis of data from the whole period indicated that the ADG showed a quadratic relationship (R^2 = 41.26% and P = 0.04) between the diets, with a tendency for a higher growth rate with the intermediate diets combining soybean oilcake and sweet lupins (Figure 3.5). Regression analysis of the ADG during the starter phase revealed that a cubic function fitted the data best (R^2 = 51.09% and P = 0.04), with 75LD having the highest ADG and 100LD the lowest.

It must be noted that although some of the regression models are significant ($P < 0.05$), the R^2 values of the regression models presented in Table 3.10 give an indication of how close the data lie to the fitted regression line. As the R^2 values were not very high, the regression models did not explain a high percentage of the variation in the data. The highest R^2 value found was 62.82%. Thus, the regression functions do not describe the effect that the treatments have on the production traits very accurately.

Table 3.9. *P values from the ANOVA for the treatment differences, polynomial contrasts and LSMEANS of the treatment diets for the production traits measured over all the phases and within each phase of the trial (starter, grower and finisher) separately*

Phase	Overall				Starter				Grower				Finisher			
Traits	EW[1]	DMI[2]	ADG[3]	FCR[4]	EW[1]	DMI[2]	ADG[3]	FCR[4]	EW[1]	DMI[2]	ADG[3]	FCR[4]	EW[1]	DMI[2]	ADG[3]	FCR[4]
P values Diet																
	0.34	0.70	0.21	0.96	0.07	0.52	0.09	0.47	0.25	0.88	0.44	0.98	0.85	0.84	0.98	0.73
P values Polynomials																
Linear	0.56	0.40	0.22	0.65	0.36	0.48	0.34	0.22	0.14	0.36	0.18	0.85	0.86	0.52	0.99	0.62
Quadratic	0.06	0.38	0.04	0.91	0.13	0.38	0.25	0.94	0.15	0.94	0.31	0.59	0.40	0.56	0.84	0.44
Cubic	0.70	0.48	0.76	0.79	0.01	0.22	0.02	0.35	0.46	0.77	0.67	0.77	0.69	0.49	0.85	0.90
Quartic	0.83	1.00	0.99	0.60	0.46	0.82	0.57	0.31	0.55	0.69	0.42	0.99	0.64	0.72	0.56	0.32
LSMEANS of treatment diets																
0L	87.5	1874.1	324.2	5.62	41.9	1104.3	405.3	2.65	68.0	1938.4	324.4	6.09	89.8	2580.4	279.6	9.25
25L	92.3	1907.4	351.4	5.61	38.4	1020.9	362.6	2.91	72.9	2019.8	359.6	5.59	93.2	2871.2	292.0	9.81
50L	94.6	2115.4	368.7	5.91	41.0	1233.9	390.5	3.33	75.5	2174.5	402.0	5.57	92.1	2968.4	262.0	12.68
75L	95.2	2227.7	368.3	5.58	45.3	1317.1	437.3	2.75	72.9	2076.6	369.7	0.76	92.3	2761.0	283.4	10.12
100L	88.9	1978.1	344.6	6.01	35.9	1095.1	328.7	3.57	73.6	2326.7	385.2	5.81	90.9	2894.6	283.2	10.61

[1]End weight (kg)

[2]Dry matter intake (g/bird/day)

[3]Average daily gain (g/bird/day)

[4]Feed conversion ratio (kg feed/kg weight gain)

Table 3.10. *Equations of the polynomial regressions fitted to the production trait trends as a result of change in the sweet lupin inclusion levels in the diets of slaughter ostriches over all phases and within each phase of the trial (starter, grower and finisher) separately.*

Trait	Phase	Function	Equation	R^2 (%)	*P* model
End Weight	All Phases	Quadratic	$y = -1.6864x^2 + 10.704x + 78.124$	38.1	0.14
	Starter	Cubic	$y = -0.0126x^3 + 0.3492x^2 - 2.2282x + 41.911$	62.15	0.03
	Grower	Linear	$y = 0.1529x + 70.28$	59.44	0.005
	Finisher	Linear	$y = 0.0694x + 91.476$	62.82	0.003
Dry Matter Intake (DMI)	All Phases	Linear	$y = 55.22x + 1854.8$	23.64	0.2
	Starter	Linear	$y = 6.03x + 1093.9$	18.75	0.29
	Grower	Linear	$y = 10.98x + 1942.7$	14.47	0.39
	Finisher	Linear	$y = 6.92x + 2723.4$	23.82	0.2
Average Daily Gain (ADG)	All Phases	Quadratic	$y = -8.53x^2 + 56.93x + 274.48$	41.26	0.04
	Starter	Cubic	$y = -0.1506x^3 + 4.19x^2 - 27.49x + 407.01$	51.09	0.04
	Grower	Linear	$y = 1.76x + 341.81$	14.84	0.16
	Finisher	Linear	$y = -0.0224x + 280.34$	0.00	0.99
Feed Conversion Ratio (FCR)	All Phases	Linear	$y = 0.0749x + 5.5216$	2.01	0.61
	Starter	Linear	$y = 0.0335x + 2.7076$	12.62	0.19
	Grower	Linear	$y = -0.0052x + 5.8418$	0.36	0.83
	Finisher	Linear	$y = 0.0457x + 9.8867$	2.21	0.6

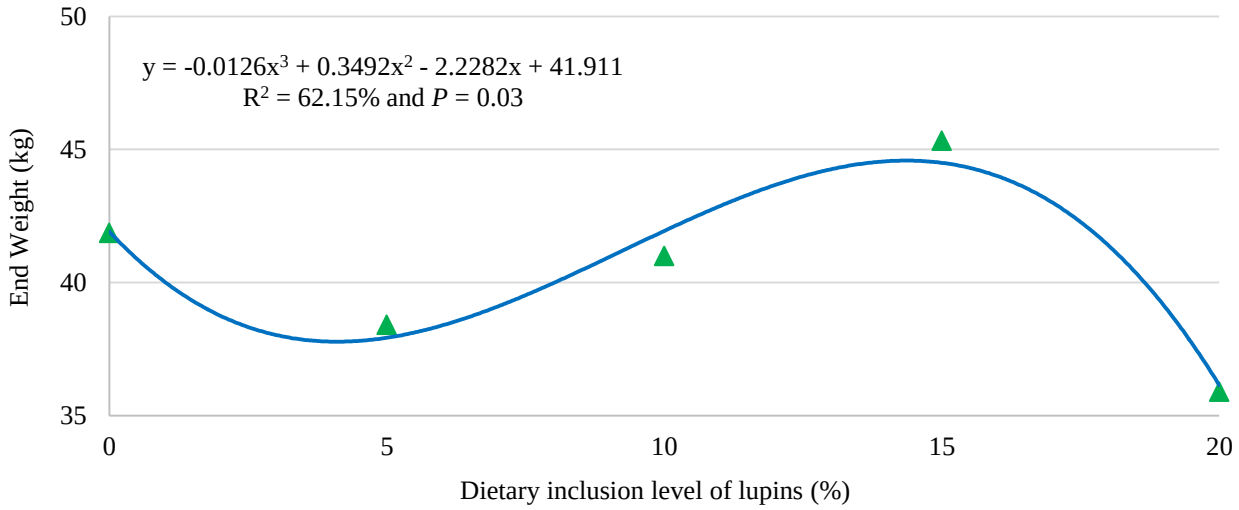

Figure 3.2. *Cubic function fitted to the end weights of starter phase slaughter ostriches fed diets with increasing sweet lupin inclusion levels (%)*

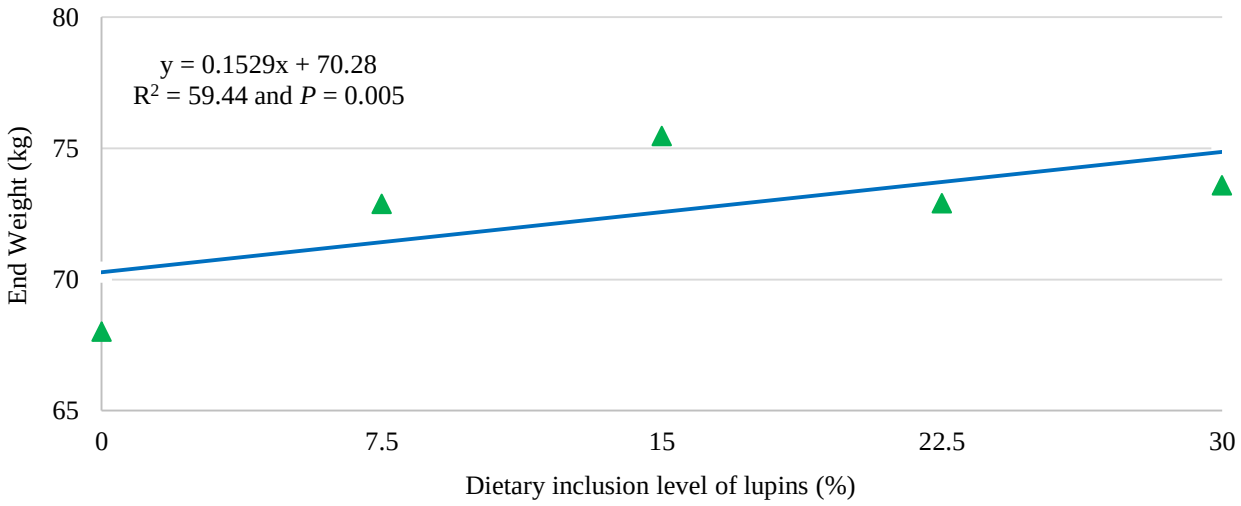

Figure 3.3. *Linear function fitted to the end weights of grower phase slaughter ostriches fed diets with increasing sweet lupin inclusion levels (%)*

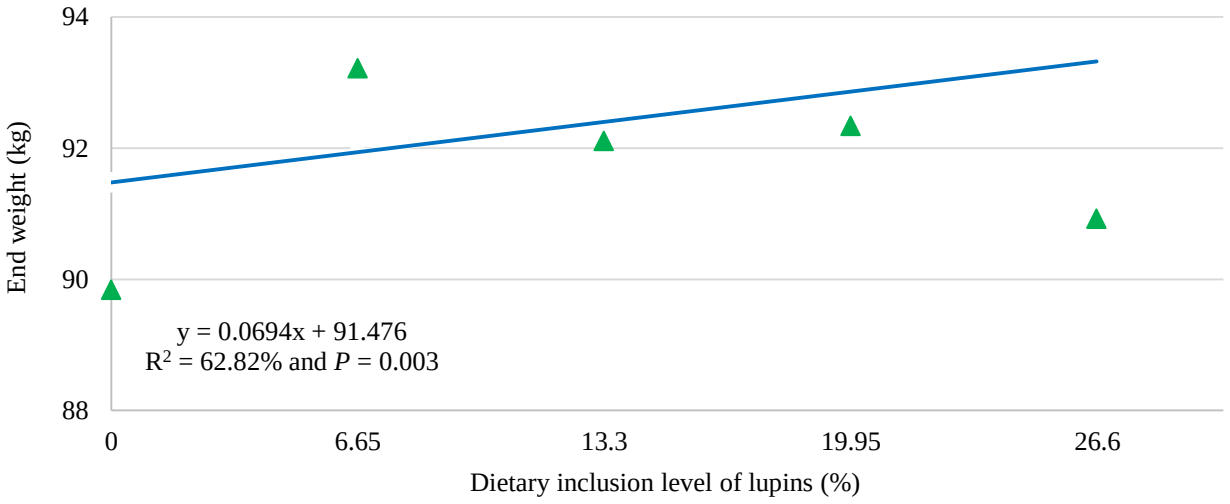

Figure 3.4. *Quadratic function fitted to the end weights of finisher phase slaughter ostriches fed diets with increasing sweet lupin inclusion levels (%)*

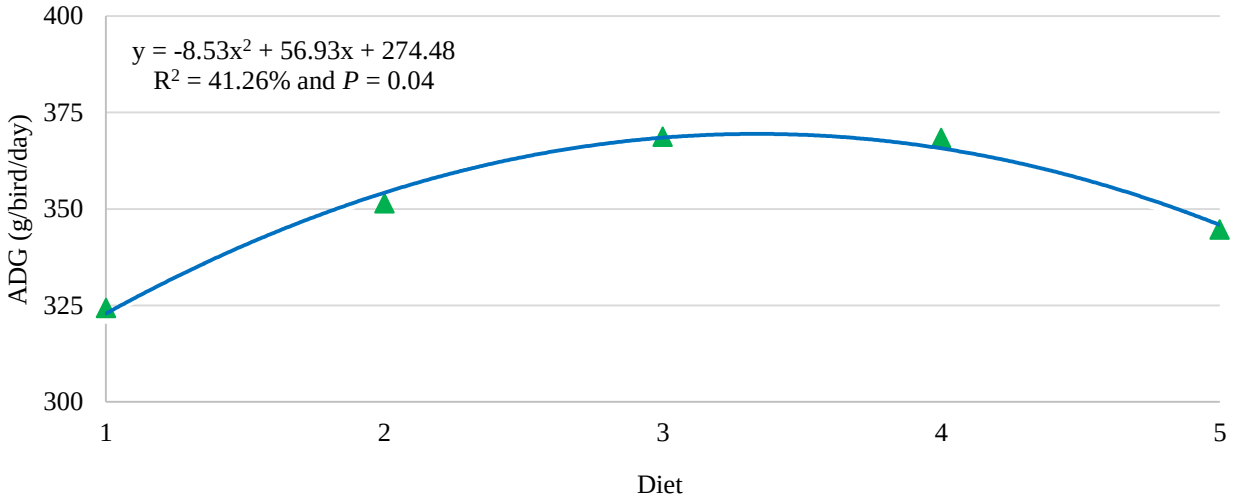

Figure 3.5. *Quadratic function fitted to the ADG over the entire growth period of slaughter ostriches fed diets with increasing sweet lupin inclusion levels (%)*

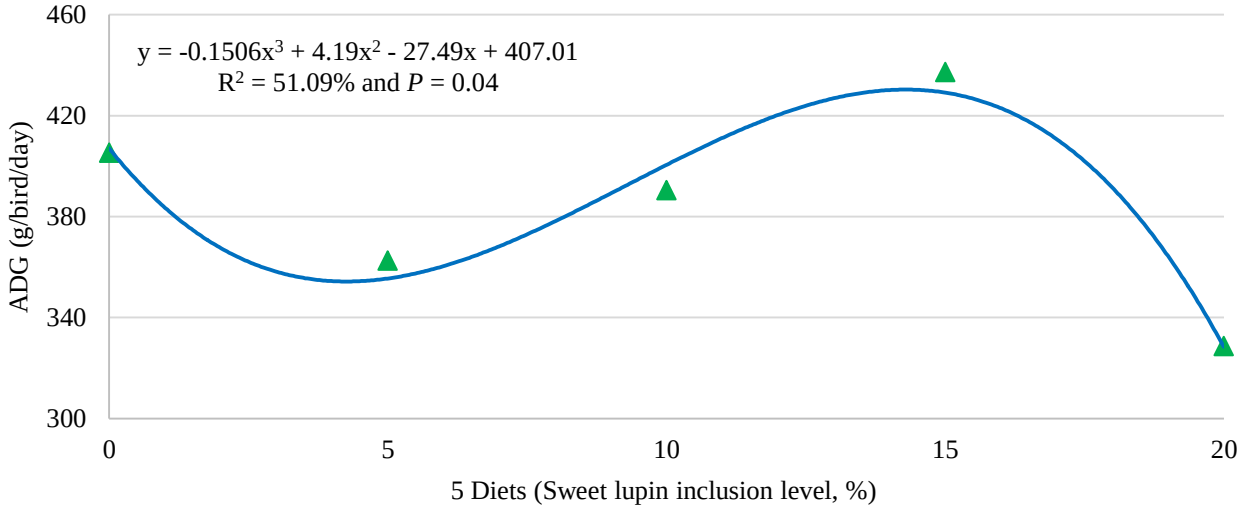

Figure 3.6. *Cubic function fitted to the ADG of starter phase slaughter ostriches fed diets with increasing sweet lupin inclusion levels (%)*

No differences were found between the treatment diets in terms of the final weights (P = 0.36) and the slaughter weights of the ostriches at the abattoir (P = 0.32) (Table 3.11). There was a tendency (P = 0.08) for the cold carcass weights to differ, with birds on the 50LD and 75LD having the heaviest weights of 44.3 ± 1.04 kg and 43.3 ± 1.04 kg respectively, which did not differ. Birds fed the 0LD and 25LD had lighter carcasses (41.7 ± 1.04 kg and 42.4 ± 1.04 kg respectively), which did not differ from ostriches on the 50LD and 75LD or from the birds fed the 100LD, which had the

lightest carcasses at 39.6 ± 1.04 kg. However, the cold carcass weights of birds fed the 50LD and 75LD differed ($P < 0.05$) from those fed the 100LD, weighing 10.6% more. With regards to the dressing percentage, no differences ($P = 0.11$) were observed between the treatments (Table 3.11). Some variation ($P = 0.05$) was however found in the right thigh weights, with the thighs of birds receiving the 50LD weighing 12.3% more ($P < 0.05$) than that of those receiving the 100LD. The right thigh weights of the birds on the 0LD (15.3 ± 0.38 kg), 25LD (15.5 ± 0.38 kg) and 75LD (16.0 ± 0.38 kg) had intermediate values. Furthermore, no differences were found in the combined thigh weights as a percentage of the carcass ($P = 0.37$), the weights of the abdominal fat pads ($P = 0.15$) or the big drum muscles ($P = 0.16$), or for the big drum muscles when expressed as a percentage of the right thigh ($P = 0.59$).

Table 3.11. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin inclusion levels on the measured slaughter traits of slaughter ostriches at the end of the experimental period*

Traits	Diets				
	0LD	25LD	50LD	75LD	100LD
Final weight (kg)	86.9 ± 2.91	92.8 ± 2.91	94.8 ± 2.91	93.6 ± 2.91	89.8 ± 2.91
Slaughter weight (kg)	81.5 ± 2.91	85.5 ± 2.91	88.6 ± 2.91	87.7 ± 2.91	82.2 ± 2.91
Cold carcass weight (kg)	41.7^{ab} ± 1.04	42.4^{ab} ± 1.04	44.3^{a} ± 1.04	43.3^{a} ± 1.04	39.6^{b} ± 1.04
Dressing percentage (%)	51.7 ± 0.79	49.8 ± 0.79	50.1 ± 0.79	49.5 ± 0.79	48.3 ± 0.79
Right thigh weight (kg)	15.3^{ab} ± 0.38	15.5^{abc} ± 0.38	16.4^{c} ± 0.38	16.0^{cb} ± 0.38	14.6^{a} ± 0.38
Thigh (both) weight as percentage of carcass (%)	73.2 ± 0.42	73.1 ± 0.42	74.1 ± 0.42	73.6 ± 0.42	74.0 ± 0.42
Abdominal fat pad weight (kg)	4.05 ± 0.39	4.78 ± 0.39	5.26 ± 0.39	5.42 ± 0.39	4.44 ± 0.39
M. gastrocnemius (big drum) weight (kg)	0.88 ± 0.03	0.91 ± 0.03	0.97 ± 0.03	0.90 ± 0.03	0.85 ± 0.03
M. gastrocnemius percentage of right thigh (%)	5.78 ± 0.11	5.84 ± 0.11	5.87 ± 0.11	5.62 ± 0.11	5.82 ± 0.11

[a,b,c] Row means with different superscripts differ significantly ($P < 0.05$)

3.4. DISCUSSION

According to Dalle Zotte *et al*. (2013), all of the nutrient requirements of an animal must be met in order to successfully rear any livestock species. Up until 1995, ostrich diet formulations were largely based on energy values for poultry ingredients as information on ME values derived for ostriches were scarce (Angel, 1996). However, computer-based mathematical simulation models have since been developed by Gous and Brand (2008), and are a useful tool in determining least-cost diet formulations that can help feed manufacturers improve the competitiveness of the ostrich industry. These models have been updated to take into account the effects of different energy and protein levels on ostrich production (Carstens, 2013).

Lupins are cheaper than other protein sources, making them widely used in livestock feeds (Edwards and van Barneveld, 1998). Only soybeans, which are a more popular protein source, can compete with lupins in this respect. However, lupins have a fibrous seed coat which contributes to their higher NDF content (201.1% more NDF than soybean oilcake meal) and affects the digestibility of the meal (McDonald *et al*., 2011). This is a major concern for young monogastric animals (Brand, 1996), but the ostrich – a hindgut fermenter – has the ability to digest fibrous material (Brand and Olivier, 2011). This allows ostriches to utilise a greater proportion of the available energy in the feed than poultry and pigs (Brand *et al.*, 2000c; Kruger, 2007). Swart (1988) found that ostriches could possibly absorb between 12 – 76% of their energy in the form of volatile fatty acids, which are the end products of fibre digestion in the large intestine. It is, however, important to remember that ostrich chicks are not able to utilise fibre efficiently before approximately three months of age, as their ability to digest crude fibre only develops thereafter (Angel, 1996). This gives rise to the need for different feeding phases at specific growth stages when ostriches are reared intensively. Swart (1988) also described the ability of the ostrich to utilise low quality raw materials, showing that they can effectively digest up to 66% of the hemicellulose and 38% of the cellulose in a feed.

Currently, lupin seed meal serves as an economic alternative to soybean meal and rapeseeds thanks to the low-alkaloid sweet varieties that are available (Ewing, 1997). As mentioned, lupins contain alkaloids that can be toxic to the animal. However, the seeds of *L. angustifolius* cultivars bred for low levels of alkaloids maintain a low and relatively stable alkaloid content (Brand and Brandt, 2000). In order to prevent the risk of toxicity, only alkaloid levels lower than 0.06% are deemed suitable for animal feeds (McDonald *et al*., 2011). In this study, no alkaloids were found in the respective feed samples as analysed using the method described by Boschin *et al*. (2008) at a detection limit of 100 ng/ml. The estimated total alkaloid content per diet, calculated from the total alkaloid content of the sweet lupin cultivar found by Brand and Smith (2016), were also below 0.06%. The use of the provided diet formulations (Table 3.4 – 3.6) with prolonged intakes should

therefore not present any risk of toxicity for the birds and can therefore be regarded as safe. They should also not influence the feed preference of the birds, which can be a problem with high alkaloid levels as they decrease the palatability of a feed (Smith, 2005).

Long term feed intake is primarily influenced by the energy content of the diet, as an animal will eat in order to meet its energy requirements (Forbes and Shariatmadari, 1994). Throughout the trial period, the experimental diets were formulated to meet the nutrient requirements of the birds and have equal ME values; any differences found in the nutrient components are deemed to be too small to have had a significant effect on the DMI of the birds. Lupins contain a relatively high protein content (32%), although they have a limited biological value due to their low methionine and proportionally low lysine content in comparison to other legumes (Ewing, 1997; Edwards and van Barneveld, 1998). These shortfalls can be supplemented by including other protein sources in the diet or by using synthetic amino acids. Synthetic methionine and lysine were thus included in the respective trial diets where necessary.

The Gompertz equations fitted to the mean weights of the birds per paddock for each diet, over the course of the experimental period, revealed sigmoidal growth curves for all the diets. As mentioned, the growth stage of the bird will determine its nutrient requirements, because as the bird grows, its body composition in terms of the protein:fat ratio changes (Brand and Olivier, 2011). There were in fact no differences in the general growth patterns of the birds (three different phases: initial self-accelerating, intermediate linear and self-decelerating) between the five dietary treatments. It can therefore be concluded that the composition of the diets did not impede the growth of the ostriches.

Brand and Gous (2006) stated that the nutrient composition of the feed will determine the production rate of growing ostriches. In terms of the production traits (end weight, DMI, ADG and FCR) of these slaughter ostriches, results were within the bounds determined by previous intensive slaughter ostrich production studies (Brand *et al*., 2000b; Brand *et al*., 2000d; Brand *et al*., 2004b; Brand *et al*., 2004c; Dalle Zotte *et al*., 2013 and Brand *et al*., 2014). This indicates that the production traits for all the diets resembled that of the production norms for slaughter ostriches.

In this study, a tendency (P = 0.09) was found for the ADG of the starter phase to vary between the diets, with the birds fed the 75LD tending to have a higher ADG than the birds fed the 100LD, although no differences were observed for the DMI and FCR. The DMI values were similar to those of Brand *et al*. (2000b), who evaluated the effects of three different crude fibre levels on the production performance of slaughter ostriches. In contrast, Brand *et al*. (2000b) found differences in the ADG and FCR. The greater number of treatment diets used in the sweet lupin trial compared to Brand *et al*. (2000b), as well as the knowledge gained over the years in terms of understanding the nutritional requirements of the ostrich since the work by Brand *et al*. (2000b),

could well be responsible for the difference in the trends. However, Brand *et al*. (2014) found differences in all three production traits when evaluating low, medium and high energy diets.

During the grower phase of the sweet lupin trial, no differences were found for the DMI, ADG or FCR. These results are supported by Brand *et al*. (2004b) and Brand *et al*. (2004c), who also found no differences in the production traits when feeding different protein levels. However, studies on the effects of different dietary energy levels by Brand *et al*. (2004b) and Brand *et al*. (2004c) showed differences in each trait between the different treatment diets. Brand *et al*. (2014) found no differences in the DMI of the birds, which supports the results found in the sweet lupin trial, but the results are inconsistent in terms of the ADG and FCR, where differences were observed. However, Brand *et al*. (2000b) found no differences in the FCR of the birds, but differences were observed for the DMI and ADG (about 1569.0 and 358.3 g/bird/day respectively) of the birds. Relative to the sweet lupin trial, Brand *et al*. (2000b) observed lower intakes but still achieved similar growth rates and thus better FCRs, ranging between 3.80 – 5.08 kg feed/kg weight gain.

Regarding the finisher phase of the sweet lupin trial, once again no differences were observed between the treatment diets. The same trend was followed for the production traits in the finisher phase of Brand *et al*. (2000b) as for the grower phase. The DMI and ADG revealed differences between the three treatment diets, but no differences were observed for the FCR, supporting the results found in the sweet lupin trial. The results of the protein trial by Brand *et al*. (2004b) partly support the findings of the sweet lupin trial, with no differences observed in the ADG or FCR of the birds. However, the DMI of the birds was higher than that found in the current trial (3306.0 *versus* 2815.1 g/bird/day). This was unexpected as the birds in the sweet lupin trial were slaughtered at 11 months of age and not 12 as in the protein trial of Brand *et al*. (2004b).

Brand *et al*. (2000d) found no differences in the DMI, ADG or FCR when feeding ostriches diets differing in protein content (13, 15 and 17% protein) from four to 11 months of age. These findings support the findings of the sweet lupin trial, where no differences were observed in the production traits with regards to the whole trial period. Brand *et al*. (2000d) also fed different energy levels (9, 10.5 and 11 MJ/kg ME), which also did not result in any differences in the ADG (approximately 302.3 g/bird/day) of the birds, but differences were observed for the DMI (2647 g/bird/day) and FCR (7.68 kg fee/kg weight gain) (Brand *et al*., 2000d). Contradictory to these findings, Dalle Zotte *et al*. (2013) found differences in the ADG of ostriches fed 0, 3, 6, 9 and 12% dietary levels of cottonseed oilcake meal, but no differences for the DMI and FCR. However, guidelines for feeding and managing ostriches between six and 13 months of age recommend a DMI ranging from 1.1 and 2.9 kg/bird/day, an ADG of 200 – 340 g/bird/day and a FCR of between 5 and 15 kg feed/kg weight gain (Brand, 2014). The results found in the respective studies are thus

in agreement with the guidelines provided by Brand (2014). It is however important to remember that as the bird ages, the ADG decreases (Brand and Gous, 2006), a trend also observed in these studies.

Regarding the slaughter traits of the birds, the dietary treatments did not influence growth or production. This can somehow be expected since the ostrich has the ability to digest fibrous material the higher dietary sweet lupin inclusion could also be utilised. The values obtained in the sweet lupin trial resembled the findings of Brand *et al.* (2004a), Hoffman *et al.* (2007) and Viviers (2015), who also evaluated the slaughter traits of ostriches in growth studies. The birds in the sweet lupin trial were slaughtered at 11 months of age rather than 12 – 14 months of age, as was done in the cited studies. They therefore attained a lower mean final live weight. The cold carcass weight and right thigh weight in the current study followed the same trend as the mean final weight (Kritzinger, 2011), with the birds slaughtered at an early age (11 months of age) having a lighter cold carcass and consequently lighter right thighs. Therefore, the big drum muscles of the birds in the sweet lupin trial were also lighter than those found by Viviers (2015). However, the mean dressing percentage (49.9%) fell within the range reported for previous studies, with 45.1% for the energy trial by Viviers (2015) and 51.1% for Hoffman *et al.* (2007). The yield of both thighs expressed as a percentage of the carcass was similar to that reported by Viviers (2015).

Brand *et al.* (2004a) found a mean abdominal fat pad weight of about 6.4 kg, while Viviers (2015) reported a lower value of 5.4 kg. However, in both aforementioned studies the birds were slaughtered at 12 months of age, so the lower fat pad weight (mean value of 4.8 kg) of the birds in the sweet lupin trial is to be expected. Differences in the abdominal fat pad weight were reported in the energy trial by Brand *et al.* (2004a), but Viviers (2015) and Brand *et al.* (2004a) (protein trial) did not observe any differences, which is in agreement with the results of this study. Hoffman (2005) postulated that any extra energy in the diet is stored in the abdominal fat pad and not in the intra-muscular fat reserves.

Overall it can be concluded that sweet lupins can successfully be included in the diets of slaughter ostriches up to 15% (75LD) in the starter phase and 30% (100LD) in the grower and finisher phases. Similar results were achieved for the production and slaughter traits measured as reported for previously performed studies, suggesting that the sweet lupin inclusion did not have any negative impact on the birds. The fact that there were overall no differences in the production traits or measured slaughter traits between the treatments during the different feeding phases is satisfactory. However, some variation was found in the end weight and ADG of the starter phase and for the cold carcass and right thigh weights. Nonetheless, it appears that sweet lupins can replace soybean oilcake meal in the diets of slaughter ostriches up to the dietary sweet lupin

inclusion levels evaluated in this study without any detrimental effect on any of the production and slaughter traits, as in this study the birds showed acceptable growth performance.

When these findings are compared to results obtained and recommendations made by other researchers, it is clear that the ostrich is able to consume and utilise this raw material at higher inclusion levels than other monogastric animals. In The Feeds Directory by Ewing (1997), the recommended inclusion rates of lupin flakes/meal per species are as follows: 0% in creep, weaner and grower diets of pigs, but for finisher and sow diets 7.5% is recommended; in poultry diets 0% inclusion is advisable in chick diets, 5% in broiler diets and 7.5% in breeder and layer diets.

According to McDonald *et al*. (2011), the maximum lupin inclusion levels in the diets of adult poultry and pigs is 10% and for growing pigs and broilers, 5%. Brand *et al*. (1995) noted that lupins can be included up to 20% in the diets of growing-finishing pigs as a replacement for fishmeal. However, a lower intake and growth rate can be expected due to the higher fibre content of lupins. Kim *et al*. (2012) reported that the inclusion level of lupins should be limited to 5 – 10% in commercial piglet weaner diets as lupins consist of approximately 25% seed coat (mostly insoluble fibre), while its kernel contains about 30% cell wall materials (polygalacturonans), which cause a dilution effect on the nutritional density of the balanced feed. However, there is a general perception that lupins can be included at levels higher than 15% in the diet of weaner pigs, provided that the indigestible hull is removed.

Lupins can thus be included in the diets of slaughter ostriches at higher inclusion levels than for poultry and pigs, without any concern regarding the digestibility of the feedstuff and treatment prior to feeding. This can be attributed to the hindgut fermentation ability of the ostrich which enables them to utilise fibrous materials more efficiently. This is also confirmed by Brand *et al*. (2000b), who found that ostriches can perform well on high fibre diets, without effecting their production negatively.

3.5. CONCLUSION

From this study it is concluded that sweet lupins can be included in the diets of slaughter ostriches at up to 15% (75LD) in starter diets and 30% (100LD) in grower and finisher diets to replace soybean oilcake meal, without any significant detrimental effect on the production and slaughter traits. The hindgut fermentation ability of ostriches most likely enables them to utilise sweet lupins very effectively, despite their higher fibre content than soybean oilcake meal. These results will contribute to the limited knowledge on ostrich nutrition and will be incorporated in the mathematical optimisation model developed by Gous and Brand (2008) to predict feed intake and other production traits more accurately. These findings may also assist in creating a potential

market for locally produced protein sources such as lupins and broaden our knowledge with regard to the potential of this raw material as a feed ingredient for animals.

3.6. REFERENCES

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CHAPTER 4

THE EFFECT OF VARYING SWEET LUPIN DIETARY INCLUSION LEVELS ON THE FEATHER CLASSES, LEATHER TRAITS AND MEAT COMPOSITION OF FEEDLOT OSTRICHES

Abstract

1. The South African ostrich industry was founded on the selection and breeding of ostriches for their feather quality; however, it has since become a producer of feathers, leather and meat. Despite the progress that has been made in optimising production practices, additional information is still necessary about the value of different raw materials as feed to ensure cost-efficient production.

2. This study aimed to determine the effects of the gradual replacement of soybean oilcake meal with sweet lupin (*Lupinus angustifolius*) seed in the diet of feedlot ostriches by evaluating the three economically important products. The chicks received a standard commercial pre-starter ostrich diet, with the trial utilising 141 ostrich chicks and starting at the onset of the starter phase (83 days post-hatching).

3. Five iso-nutritional diets were formulated for each feeding phase (starter, grower and finisher) according to specifications set out for each feeding phase. Within each feeding phase a control diet (diet 1, 0% lupin diet; LD) was formulated using soybean oilcake meal as the sole protein source and diet 5 (100LD) was formulated to include the maximum amount of sweet lupins according to the specifications for the specific species and feeding phase. Maximum amount of sweet lupins included in the 100LD therefore differs between the three feeding phases. The remaining three diets were formulated by mixing the diets to determine the gradual increase in lupins by replacing soybean oilcake meal in the diets from the 0LD up to the 100LD: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) and 0:100 (100LD). There were three replications per treatment, resulting in 15 groups of birds. Feed and water were supplied *ad libitum*.

4. No differences were found for the moisture, crude protein and ash content of the meat. However, the intra-muscular fat content was influenced (P = 0.03) by the sweet lupin content of the diet. No differences were found for any of the feather classes measured. Regarding the leather traits, the treatment diets had no effect on the crust sizes, leather grades, nodule diameters and nodule densities. Differences were observed for leather thickness and pinhole number. Birds receiving the 25LD and 50LD differed from birds on the 100LD, having 66.7% more pinholes on their leather; however, birds fed the 0LD and 75LD did not differ from the 25LD, 50LD or 100LD.

5. It can be concluded that the sweet lupin inclusion levels evaluated in this study had little influence on the leather traits, the meat composition of the birds measured and the feather classes.

4.1. INTRODUCTION

Between 1838 and 1866 the South African ostrich industry became established, with the primary source of income being the export of ostrich feathers to Europe (de Mosenthal and Harting, 1897). In 1913 ostrich feathers ranked as the fourth largest South African export product after gold, diamonds and wool (Jorgensen, 2014; NAMC, 2010). The feather market collapsed after the First World War, but after the Second World War there was a revival in the use of ostrich feathers for dusters and fashion and the industry slowly recovered by expanding their trade to also include leather (Deeming, 1999; Anon, 2004; NAMC, 2010). The value of ostriches as a potential source of meat remained unknown until the beginning of the 1990's (Majewska *et al.* 2009). In 2000 the production of ostrich meat and the price thereof increased steeply due to the growing international consumer preference for healthier meat. In addition, the outbreak of *Bovine spongiform encephalopathy* (BSE) and *Foot-and-mouth* disease in large parts of Europe led to an increase in demand for safe alternative red meat products (Brand and Jordaan, 2011).

The modern ostrich industry thus has three economically important products: feathers, leather and meat, which are regarded as niche products (DAFF, 2014). All three of these products should be taken into account to ensure the cost-efficient production of slaughter birds (Engelbrecht, 2014). Currently 45% of the value of a slaughtered bird is derived each from the skin and meat and the remaining 10% is earned from feathers (DAFF, 2014). The ostrich industry makes a substantial contribution to the national economy, with South Africa being regarded as the world leader of ostrich products, supplying between 75 – 85% of the global market share (Brand and Jordaan, 2011; DAFF, 2014; Kleyn, P., Pers. Comm., South African Ostrich Business Chamber, PO Box 952, Oudtshoorn, 6620, South Africa, 15 Sept. 2016).

Ostrich feathers are exported to almost every continent, from South America to Asia. They are not only a popular fashion product and used for carnivals, but their unique ability to generate static electricity makes them popular industrial and household dusters (van Zyl, 2001; DAFF, 2014). Ostrich leather is regarded as a luxury product and of all the exotic leathers, it is one of the most attractive, supple and durable (Engelbrecht, 2014; NAMC, 2010). The presence of the feather follicles or nodules on ostrich leather gives it its unique appearance, while the nodule diameter and shape contribute to the quality of the leather (Meyer *et al*., 2004; Engelbrecht, 2014).

The value of ostrich meat was only discovered recently and its health benefits, namely a low sodium content, favourable fatty acid profile, low intra-muscular fat and cholesterol content and high iron and vitamin E content, has resulted in consumers paying more attention to the ostrich as a meat producer (Mellett, 1992; Sales and Oliver-Lyons, 1996; Majewska *et al*., 2009; Poławska *et al*., 2011). However, little is known about the effect of production systems, in particular feed source, on the nutritional quality of this 'new' alternative red meat (Girolami *et al*., 2003; Poławska

et al., 2011). Information on ostrich meat has become essential, as the modern consumer wants to be informed of the nutritional value of the food they consume (Sales *et al*., 1996; Horbańczuk and Sales, 1998; Hoffman *et al*., 2005).

It remains a challenge to produce slaughter ostriches and their end products with good profit margins and there is presently huge pressure on ostrich producers to farm as cost-effectively as possible. Continuous research is therefore necessary to ensure the profitability and relevance of the ostrich industry in the agricultural sector. Factors such as high input costs, exchange rates and market trends should be considered. Feed costs make up the largest proportion of the input costs of an intensive ostrich production unit (*ca*. 75%) (Brand *et al*., 2002; Brand and Jordaan, 2011). The current widespread droughts, high costs of raw materials and the growing need for protein in human and animal diets globally (Brand *et al*., 2004a; AFMA, 2015) emphasises the need to investigate the possibility of alternative protein sources for animal feed, such as sweet lupins (*Lupinus angustifolius*). If the cost associated with nutrition can be reduced, it will have a major impact on the profitability of a commercial ostrich production enterprise.

The aim of this study was to assess the influence of the gradual replacement of soybean oilcake meal with sweet lupin seed in the diets of feedlot ostriches on the major products: feather classes, leather traits (crust sizes, leather thickness, leather grades, nodule diameters and nodule and pinhole densities) and the nutritional value of the meat (*Muscularis gastrocnemius*).

4.2. MATERIALS AND METHODS

The birds used in this study were the same birds as described in chapter 3. The full description of the experimental design of the trial, the different dietary treatments per feeding phase, the management practices and the lead up to the slaughter date and management thereof can thus be found in chapter 3.

4.2.1. Dietary treatment

Briefly, five iso-nutritional diets were formulated each for the starter, grower and finisher phases using Mixit2+ software (Agricultural Software Consultants Inc., San Diego, USA), according to specifications predicted by a model developed by Gous and Brand (2008) for the different feeding phases, i.e. starter, grower and finisher. Within each feeding phase a control diet, diet 1 (0% lupin diet; LD), was formulated using soybean oilcake meal as the sole protein source. Thereafter diet 5 (100LD) was formulated to include the maximum amount of sweet lupins according to the specifications for the specific species and the feeding phase. The maximum amount of sweet lupins included in the 100LD therefore differs between the three feeding phases. The remaining three diets were formulated by mixing the diets to determine the increase in sweet lupins

in the diets from the 0LD up to the 100LD. Soybean oilcake meal was thus gradually replaced with sweet lupins in the following ratios: 100:0 (0LD), 75:25 (25LD), 50:50 (50LD), 25:75 (75LD) and 0:100 (100DL). The raw material composition of the soybean oilcake meal and sweet lupin used in the diet formulations and nutrient composition of the different dietary treatments can be found in Tables 3.4 – 3.6 in chapter 3. The respective diets, as well as clean and fresh water, were available *ad libitum* to the birds throughout the different feeding phases.

4.2.2. Slaughtering procedures

The birds were slaughtered at *ca*. 11 months of age at a registered abattoir, Mosstrich, in Mossel Bay. The slaughtering procedures used by the abattoir staff were similar to those described by Hoffman (2012). After exsanguination, the feathers of each bird were harvested, kept separately in individually marked bags and transported to the feather department of Klein Karoo International (KKI) Limited in Oudtshoorn for drying and processing. After skinning, the individual skins of each bird were marked and transported to the premises of Southern Cape Ostrich Tanning (SCOT) for further processing. The carcasses were stored in the cold storage facilities (0 – 2°C) for 18 hours before deboning of the birds commenced. The weight of the big drum muscle (*M. gastrocnemius*) of the right thigh of each ostrich was recorded and selected samples were collected and transported back to Stellenbosch University for further chemical analysis.

4.2.3. Feather harvesting, processing and classification

It is normal industry practice to clip the ripe feathers from ostriches of six months of age or a live weight of 60 kg or more in order to ensure a good feather crop at slaughter (12 months of age). The harvesting of immature or green/blood feathers is not permitted due to welfare concerns (Engelbrecht, 2014), and their plucking results in the stretching of the follicles, which negatively impacts leather quality (Engelbrecht *et al.*, 2009). In this trial, the birds were not clipped or plucked as they still had blood feathers at six months of age. In addition, as the birds were slaughtered at approximately 11 months of age rather than 12 months of age, there would thus not have been adequate time for the new feathers to grow and be ripe at slaughter. In the month after feather harvesting, ostriches sometimes show a slight decline in weight. As the birds would have experienced a decline in weight if the feathers were clipped at six months of age, the cold weather conditions during May would have worsened the situation and an undesirable growth curve would have been experienced.

Once at the Klein Karoo International feather department, the feathers of each bird were weighed and placed into a large oven set at 50°C. The feathers were dried for a period of 48 hours and during the last half hour the temperature of the oven was increased to 70°C for sterilisation. As

the individual feather batches were removed from the oven, they were weighed again in order to calculate the amount of moisture lost during the drying process. The feather batches per bird were then separated into their economically important classes, namely 'marketable and unmarketable feathers', 'chick wing', 'drab silver floss', 'chick body short', 'chick blondene floss', 'chick tail' and 'ruggies' (back feathers), and graded by qualified graders. The description of the different feather classes can be found in Table 4.1. The weight of each separate class of feathers was recorded for statistical analysis.

Table 4.1. *Description of the different economically important feather classes found in this study*

Feather classes	Area of body and description
Unmarketable	Do not meet the grading standards set out by the industry; these feathers originate from the wings, body and tail of the bird.
Chick wing	The white plumes in the first row of prominent plumes at the edge of the wing.
Drab silver floss	A soft body feather that is found on the ventral side of the bird; they are generally shorter in length. The feather classes classified as 'drab' are normally feathers that are shorter than the wing feathers due to their location on the body.
Chick body short	The body feathers from behind the thighs to the tail.
Chick blondene floss	A soft body feather that is found on the wings of the bird and is the only soft feather on the wings.
Chick tail	A body feather found on the tail part of the bird.
Back feathers	Feathers that are found on the back of the bird.

4.2.4. Tanning process and leather grading

The tanning of the skins started soon after slaughter[1]. Once the subcutaneous fat had been removed from the raw skin by a process called hand fleshing each individual skin was assigned a marked microchip for identification along the tanning process. The removal of the fat allows the full penetration of the skin by the preservative chemicals involved in tanning as well as making the skin surface thinner and more even. A quality control check was done to determine whether any damage had occurred during the slaughtering process, before the skins were classed into weight groups. This involves weighing the skins and dividing them into the following groups: 2-3 kg, 3-4 kg, 4-5 kg, 5-6.5 kg and 6.5 kg and above. The skins were then placed in the tanning drums to undergo soaking, dehairing and liming. The pH in the drum was increased to 9.5 during the soaking process, 11.5 for the dehairing process and up to 13.5 for the liming process. The revolutions per minute (rpm) of the

[1]Due to intellectual property, specific details on the tanning process cannot be reported. However, care was taken to ensure that all skins received the same treatment.

drum were 4 rpm for 24 hours for the soaking process and for the dehairing and liming processes 22 rpm for 24 hours. Next the skins were exposed to various enzymes to break down the soluble proteins and fats still remaining in the skin, without causing any damage to the skin itself. This process is referred to as 'bating'. Thereafter the skins go through a process called 'pickling', which involves subjecting them to a salt and acid solution to prepare them for bleaching. For better penetration of the chemicals in the subsequent steps, the loose fibres from each skin were then shaved off. During the degreasing process the skins were subjected to a saponification agent specially manufactured for ostrich skins. A phase known as 're-tanning' followed. Here the style of the individual tanner becomes evident in the leather characteristics; care was taken to ensure that the same tanner worked on all the experimental skins. Due to the residual fat content, an industrial tumble drier was then used to dry the skins. The skins are also tumbled together with a cattle-hide to make the leather more soft and durable. Thereafter the dry skin size (dm^2) was measured and the skins were sorted and graded (subjective evaluation) by qualified graders. The skins were then stored in their chrome-crusted form for transport to the Elsenburg Research Institute for further data collection on a number of quality variables.

The crust size (dm^2) and grading of the leather for each bird was obtained from the tannery (SCOT, 2016). The leather thickness (mm) was measured with a Mitutoyo digital caliper (model number: CD-8" C) on the right hand side of the skin, next to and outside of the crown area as described by Engelbrecht (2013). Three separate measurements were taken to obtain an average thickness for each crust. The leather of each bird was evaluated at the five different localities, as indicated in Figure 4.1. The base diameter of five randomly selected nodules per 1 dm^2 sampling site was also measured with the Mitutoyo digital caliper and an average calculated per treatment. Furthermore, the number of nodules within the 1 dm^2 was counted and thereafter the number of pinholes (remnants from the removal of the hair follicles on the skin) in a smaller 5 cm x 5 cm area within the 1 dm^2 square area was also counted so as to determine the nodule and pinhole density per locality.

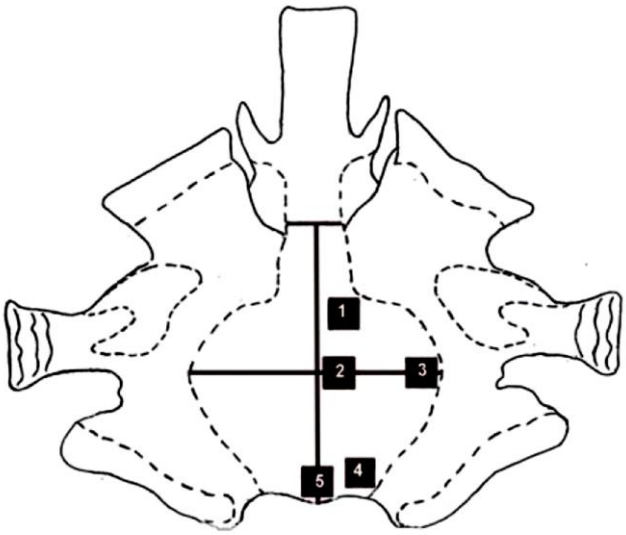

Figure 4.1. *The five different localities on an ostrich skin for which sampling and data capturing was done, viz. 1 – neck; 2 – mid-crown; 3 – upper leg; 4 – lower flank; 5 – butt (Cloete et al. 2004)*

4.2.5. Proximate analysis of the big drum muscle

Birds were sorted in ascending order according to their final live weight and ten birds per treatment were selected around the median for further chemical analysis of their meat. The big drum muscle (*M. gastrocnemius*) of the right thigh of these birds was used to perform proximate analysis once the subcutaneous fat had been removed and the meat had been homogenised. The samples were analysed using the methods of the Association of Official Analytical Chemists (AOAC) for moisture, ash, crude protein and crude fat (AOAC, 2002). The moisture content (%) was determined using method 934.01 (moisture loss on drying at 95 – 100°C) and thereafter the ash content was determined on the moisture free sample using method 942.05. Method 992.15 (Dumas combustion method) was used to determine the crude protein (*N* x 6.25) content of the meat sample and finally extraction method 920.39, chloroform/methanol (1:2 vol/vol), was used to determine the crude fat content.

4.2.6. Statistical analysis

Statistical analysis was done using SAS Enterprise Guide (Version 9.2; SAS Institute Inc., Cary, USA). The feather traits and classes, leather traits and chemical components of the meat were investigated by performing one-way analysis of variances (ANOVAs) to determine if the treatment diets had any effect over the entire experimental period. To test for normality the Shapiro-Wilk test was conducted (Shapiro and Wilk, 1965). Outliers were identified and removed prior to final analysis. The t-least significant differences were calculated on a 5% significance level. The resultant differences were deemed significant at $P < 0.05$.

4.3. RESULTS

The treatment diets had no effect on the total average dry feather weight and the total average dry feather weight marketable (Table 4.2). Similarly, no differences were observed between the diets for any of the measured feather classes (Table 4.2).

Table 4.2. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin dietary inclusion levels over the entire experimental period on the feather traits (oven dried) and classes of slaughter ostriches (11 months of age)*

Feather traits and classes (g)	Diets				
	0LD	25LD	50LD	75LD	100LD
Total dry feather weight	766.5 ± 34.8	768.1 ± 34.8	850.2 ± 34.8	787.7 ± 34.76	820.9 ± 34.76
Total dry feather weight marketable	634.9 ± 61.6	622.3 ± 61.6	705.9 ± 61.6	677.4 ± 61.57	592.5 ± 61.57
Chick wing	124.9 ± 18.2	111.7 ± 18.2	127.8 ± 18.2	136.9 ± 18.16	120.6 ± 18.16
Drab silver floss	107.6 ± 14.2	103.6 ± 14.2	124.2 ± 14.2	112.4 ± 14.16	98.1 ± 14.16
Chick body short	171.6 ± 39.2	186.4 ± 39.2	199.2 ± 39.2	143.4 ± 39.17	141.6 ± 39.17
Chick blondene floss	33.6 ± 3.59	30.8 ± 3.59	31.8 ± 3.59	36.1 ± 3.59	35.9 ± 3.59
Chick tail	32.7 ± 7.48	13.4 ± 7.48	10.9 ± 7.48	25.0 ± 7.48	24.5 ± 7.48
Back feathers	164.6 ± 25.3	176.5 ± 25.3	212.1 ± 25.3	223.8 ± 25.3	171.9 ± 25.3
Unmarketable feathers	131.5 ± 54.6	145.7 ± 54.6	144.3 ± 54.6	110.3 ± 54.6	228.4 ± 54.6

Regarding the leather traits in Table 4.3, no differences were observed between the treatment groups for the crust size, but differences (P = 0.03) were observed for leather thickness. The leather of the birds on the 25LD was significantly (9.8%) thicker than that of the birds receiving 0LD, 50LD, 75LD and 100LD.

Table 4.3. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin dietary inclusion levels on some leather traits of slaughter ostriches (11 months of age) at the end of the experimental period*

Leather traits	Diets				
	0LD	25LD	50LD	75LD	100LD
Crust size (dm^2)	136.4 ± 2.87	141.9 ± 2.87	144.2 ± 2.87	141.4 ± 2.87	136.9 ± 2.87
Thickness (mm)	0.83^a ± 0.02	0.95^b ± 0.02	0.88^a ± 0.02	0.87^a ± 0.02	0.88^a ± 0.02

^{a,b} Row means with different superscripts differ significantly ($P < 0.05$)

No differences were found with regards to the mean grade allocated to the leather of the birds in the different treatment groups, as well as for the proportions of leather allocated to the various grades (grade 1 to grade 4) between the dietary treatments (Table 4.4).

Table 4.4. *Proportion (%) ± standard error (SE) of each leather grade for the effect of sweet lupin dietary inclusion levels on the leather grades of slaughter ostriches (11 months of age) at the end of the experimental period*

Leather grades	Diets				
	0LD	25LD	50LD	75LD	100LD
Mean	2.50 ± 0.41	2.14 ± 0.41	2.27 ± 0.41	2.27 ± 0.41	2.42 ± 0.41
Grade 1 (%)	15.4 ± 10.9	30.6 ± 10.9	17.9 ± 10.9	21.4 ± 10.9	23.3 ± 10.9
Grade 2 (%)	29.9 ± 14.2	36.8 ± 14.2	50.6 ± 14.2	42.9 ± 14.2	36.7 ± 14.2
Grade 3 (%)	43.6 ± 8.84	20.0 ± 8.84	17.9 ± 8.84	22.9 ± 8.84	15.0 ± 8.84
Grade 4 (%)	11.1 ± 14.4	12.5 ± 14.4	13.7 ± 14.4	12.9 ± 14.4	25.0 ± 14.4
Total (%)	100	100	100	100	100

^{a,b} Row means with different superscripts differ significantly ($P < 0.05$)

Similarly, no differences were observed in the nodule diameters of the leather of the birds for the different dietary treatments (Table 4.5).

Table 4.5. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin inclusion levels on the nodule diameter of slaughter ostriches (11 months of age) at the end of the experimental period*

Locality	Diets				
	0LD	25LD	50LD	75LD	100LD
Locality 1 - Neck	2.92 ± 0.07	3.01 ± 0.07	3.07 ± 0.07	3.07 ± 0.07	2.95 ± 0.07
Locality 2 - Mid-crown	3.09 ± 0.07	3.00 ± 0.07	3.13 ± 0.07	3.03 ± 0.07	2.94 ± 0.07
Locality 3 - Upper leg	3.24 ± 0.07	3.32 ± 0.07	3.38 ± 0.07	3.41 ± 0.07	3.37 ± 0.07
Locality 4 - Lower flank	3.50 ± 0.07	3.57 ± 0.07	3.70 ± 0.07	3.53 ± 0.07	3.67 ± 0.07
Locality 5 - Butt	3.59 ± 0.10	3.72 ± 0.10	3.75 ± 0.10	3.61 ± 0.10	3.59 ± 0.10

In terms of the nodule and pinhole densities in Table 4.6, no differences were found between the treatment diets in the five locations measure on the leather. There did however appear to be a tendency (P = 0.08) for the dietary lupin content to have an effect on the pinhole density at locality 4 (lower flank), with the birds receiving the 25LD and 50LD having significantly more (66.7%) pinholes on their leather than the birds on the 100LD. The birds fed the 0LD and 75LD did not differ from those fed the 25LD, 50LD and 100LD in terms of the pinhole density at locality 4 (lower flank).

Table 4.6. *Least square means ± standard error (LSM ± SE) for the effect of sweet lupin dietary inclusion levels on the nodule and pinhole densities per locality of slaughter ostriches (11 months of age) at the end of the experimental period*

Traits	Diets				
	0LD	25LD	50LD	75LD	100LD
Nodule density					
Locality 1 - Neck	47.6 ± 1.53	49.2 ± 1.53	47.4 ± 1.53	49.9 ± 1.53	48.4 ± 1.53
Locality 2 - Mid-crown	59.6 ± 1.54	61.3 ± 1.54	58.4 ± 1.54	60.9 ± 1.54	60.3 ± 1.54
Locality 3 - Upper leg	24.3 ± 0.71	22.6 ± 0.71	23.8 ± 0.71	25.0 ± 0.71	24.9 ± 0.71
Locality 4 - Lower flank	36.2 ± 1.17	35.7 ± 1.17	35.1 ± 1.17	36.3 ± 1.17	35.1 ± 1.17
Locality 5 - Butt	53.1 ± 1.66	51.2 ± 1.66	49.9 ± 1.66	54.5 ± 1.66	52.8 ± 1.66
Pinhole density					
Locality 1 - Neck	32.0 ± 5.43	38.0 ± 5.43	41.4 ± 5.43	38.4 ± 5.43	30.9 ± 5.43
Locality 2 - Mid-crown	36.9 ± 6.87	48.7 ± 6.87	48.0 ± 6.87	40.8 ± 6.87	47.2 ± 6.87
Locality 3 - Upper leg	9.48 ± 2.28	11.0 ± 2.28	10.9 ± 2.28	12.4 ± 2.28	8.06 ± 2.28
Locality 4 - Lower flank	14.4^{ab} ± 2.34	21.5^{a} ± 2.34	21.4^{a} ± 2.34	17.6^{ab} ± 2.34	12.9^{b} ± 2.34
Locality 5 - Butt	52.9 ± 7.82	71.0 ± 7.82	65.2 ± 7.82	59.6 ± 7.82	53.8 ± 7.82

^{a,b} Row means with different superscripts differ significantly ($P < 0.05$)

No differences were observed in the moisture, crude protein and ash content of the meat for the different treatment groups (Table 4.7). A significant difference was however observed regarding the intra-muscular fat content. The 0LD and 100LD did not differ from the 25LD, but differed ($P < 0.05$) from the 50LD and 75LD, which in turn did not differ from each other for the intra-muscular fat content. The 25LD differed ($P < 0.05$) from the 50LD, but did not differ from the 75LD in terms of the intra-muscular fat content. The intra-muscular fat content of the meat of the birds fed the 50LD was 22.0% higher than the intra-muscular fat content of the meat of the birds receiving the 0LD and 100LD.

Table 4.7. *Least square means ± standard error (LSM ± SE) for the proximate analysis of the big drum muscle of slaughter ostriches fed diets with different sweet lupin inclusion levels (100% dry matter basis except for moisture)*

Chemical component (g/kg)	Diets				
	0LD	25LD	50LD	75LD	100LD
Moisture	765.6 ± 2.26	761.9 ± 2.26	764.3 ± 2.26	760.9 ± 2.26	766.5 ± 2.26
Crude protein	206.2 ± 2.47	210.3 ± 2.47	205.8 ± 2.47	207.9 ± 2.47	204.5 ± 2.47
Crude fat	23.5^{a} ± 1.24	24.6^{ab} ± 1.24	28.6^{c} ± 1.24	28.0^{cb} ± 1.24	23.3^{a} ± 1.24
Ash	11.0 ± 0.46	12.5 ± 0.46	11.2 ± 0.46	11.9 ± 0.46	12.0 ± 0.46

^{a,b,c} Row means with different superscripts differ significantly ($P < 0.05$)

4.4. DISCUSSION

This study is intended to contribute to the limited knowledge of ostrich nutrition (Brand *et al.* 2014), especially with regards to the effect of nutrition on the ostrich end products. All the treatment diets were formulated to be iso-nutritious throughout the different feeding phases evaluated. Over the entire experimental period the mean energy and protein value for the experimental diets were 12.9 MJ/kg feed and 158.0 g/kg respectively.

No significant differences were found for marketable feather yield between diets with different sweet lupin inclusion levels. This is in agreement with the results of Carstens (2013) and Viviers (2015), who also found that ostriches finished on diets with similar energy (7.5 – 15.5 MJ/kg feed) and protein (117.7 – 234.8 g/kg) contents also showed no significant differences in the marketable feather yields for the different treatment groups. However, their values were slightly higher than the results from this trial. This is mainly due to the differences in slaughter age of the birds between the experiments. The mean marketable feather yield reported by Carstens (2013), who compared different energy densities, was 790.0 ± 20.8 g and by Viviers (2015), who examined the effects of both energy and protein levels, was 945.5 ± 21.7 g and 983.9 ± 15.6 g respectively, compared to the 646.6 ± 20.1 g found in this trial.

On the other hand, values reported by Brand *et al*. (2004b) for energy (7.5 – 11.5 MJ/kg feed) and protein (80.0 – 160.0 g/kg) comparisons were even higher than the results from Carstens (2013), Viviers (2015) and the sweet lupin trial. For the energy trial a mean marketable feather weight of 1153.3 ± 32.8 g was obtained and for the protein trial 1152.0 ± 20.6 g. Brand *et al*. (2004b) also noted a significantly higher feather yield for the birds on the higher energy diets as well as finding that if the feathers of the ostriches were not clipped, a significant higher yield (21.2%) could be achieved (1260.0 g *versus* 1040.0 g). These results are contradictory to Carstens

(2013), who found that the quantity of marketable feathers was significantly higher (10.7%) for birds whose feathers were clipped. If the feathers are not harvested, development of the mature feathers only takes place at a later stage. According to Carstens (2013), new feather development starts directly after harvesting the initial feathers. The large amount of chick feather classes found during the grading process of the feathers of the birds in this sweet lupin trial could be due to the fact that the feathers were not harvested at six to eight months of age and that the bird grew slower during the winter time. Harvesting of feathers was not done at dix months of age as they were not ripe at this stage and there would have been insufficient time for complete regrowth before slaughter at 11 months of age had they been harvested.

No significant differences were observed in the crust size of the skins from birds that were supplied sweet lupins at various inclusion levels. The crust sizes are comparable to the values obtained by Brand *et al.* (2000) and Brand *et al*. (2004b). The mean crust size in this trial was 140.2 ± 1.51 dm^2 compared to the 136.3 ± 1.20 dm^2 and 138.7 ± 1.78 dm^2 crust sizes for the energy trials and 136.3 ± 0.88 dm^2 and 138.7 ± 0.37 dm^2 for the protein trials reported by Brand *et al.* (2000) and Brand *et al*. (2004b), respectively. In both these trials the skin surface area was significantly affected by the energy content of the diet, with the lower the energy content of diet, the smaller the skin size. However, the protein content of the diet had no significant effect. In the current study some of the lupin inclusion levels (25LD, 50LD and 75LD) resulted in larger crust sizes than those reported by Brand *et al.* (2000) and Brand *et al*. (2004b).

A significant difference was observed in the leather thickness of the treatment birds, with the birds receiving the 25LD having the thickest (0.95 ± 0.02 mm) leather. This finding corresponds with the slaughter weights of the birds as, although the differences were not significant, the birds on the 25LD had the highest slaughter weight (93.2 ± 2.15 kg). The mean values obtained for leather thickness in the energy and protein trials reported by Brand *et al*. (2004b) (0.97 ± 0.02 and 0.97 ± 0.01 mm, respectively) and Cloete *et al*. (2006a) (1.19 ± 0.01 and 1.19 ± 0.02 mm, respectively) were slightly higher than those in the current study (0.88 ± 0.02 mm). This might be due to the heavier slaughter weight of the birds from these studies.

No significant differences were observed between the diets for the leather grades; across all the dietary treatments grade 2 leathers were the most prevalent. These results are supported by Brand *et al*. (2004b) and Viviers (2015), who also reported no differences between the leather grades in trials examining the effects of both dietary energy and protein. In addition, these authors also reported grade 2 leathers to be the most common across the different treatment groups.

Nodule diameter showed no significant differences and values were similar to that obtained by Brand *et al*. (2004b) and Cloete *et al*. (2006b). Results indicate that locality on the skin has a marked effect on the average nodule diameter and density. The average nodule diameter on the neck

(3.00 ± 0.03 mm) and mid-crown (3.04 ± 0.03 mm) areas were smaller than those on the upper leg (3.34 ± 0.03 mm), lower flank (3.59 ± 0.04 mm) and butt (3.65 ± 0.03 mm) localities, where minor differences were observed. No significant differences in nodule density were found between the diets at the different localities measured. The nodule density was seen to decrease from the neck (48.5 ± 0.47) and mid-crown (60.1 ± 0.51) localities towards the upper leg (24.1 ± 0.44) and lower flank (35.7 ± 0.26) of the ostrich, while nearer to the back line (butt area) (52.3 ± 0.80) the nodule density increased. These observations correspond to work by Cloete *et al.* (2004), Cloete *et al*. (2006b) and Engelbrecht *et al.* (2009).

It is generally found that the average nodule diameter increases with a decrease in nodule density, with the exception of the butt area, which has a high density of large nodules (Engelbrecht *et al.*, 2009). The nodule density values per locality were overall slightly lower than the values obtained by Cloete *et al.* (2004) and Cloete *et al*. (2006b). Meyer *et al.* (2004) stated that skin damage and size were the main factors that determine ostrich skin and leather grading. However, nodule diameter was recently also incorporated as a grading factor. From the findings of Viviers (2015) it appears that dietary protein content may influence nodule diameter and density on some areas of the leather and this therefore warrants further research.

With regards to the pinhole density of the measured localities there was a tendency for the dietary lupin content to have an effect on pinhole density at locality 4 (lower flank), with the leather of the birds on the 25LD (21.5 ± 2.34) and 50LD (21.4 ± 2.34) having more pinholes than those receiving the 100LD (12.9 ± 2.34). Although Viviers (2015) did not observe any differences between ostriches fed different dietary energy and protein levels, the findings of this trial still fall within the range of values reported by Viviers (2015). The pinhole density values of locality 4 reported by Viviers (2015) ranged from 14.2 to 17.0 and 9.77 to 14.6 pinholes per skin for the energy and protein comparisons respectively, compared to the 12.9 – 21.5 pinholes per skin for this trial. However, it is important to remember that the number of pinholes is counted manually and it is difficult to accurately count these tiny holes in the leather. This results in large deviations around the mean and could possibly affect the end results. It is thus not possible to definitely confirm whether nutrition (energy, protein or sweet lupin inclusion levels) has an effect on pinhole density, although this does not appear to be the case (Viviers, 2015).

The results found in this study are thus comparable to the findings of previous similar studies and support previous conclusions on the effect of nutrition on ostrich production. Further research is necessary for the better understanding of the influence of nutrition on feather yield and quality. In the ostrich industry feathers contribute the smallest proportion (~10%) of the total income from a slaughter bird. According to Brand and Cloete (2015) this is why research and scientifically based selection programmes on this product have not been implemented. As early as 2006, Cloete *et al.*

(2006a) stated that the ostrich industry requires information on the influence of nutrition on ostrich leather quality. It appears that when ostriches receive balanced rations that meet the minimum standards for protein and energy levels, nutrition has a limited effect on skin quality. This statement is supported by Engelbrecht *et al*. (2009). However, Brand *et al.* (2000) indicated that a high dietary protein content effected the leather grading of the birds, with ostriches fed a high protein diet having more skin damage caused by scratches on the skins. This phenomenon is difficult to explain, but an excessive protein intake may cause the birds to be more restless than usual. This may be due to an accumulation of either ammonia or glutamate in their brains (Brand *et al.*, 2000).

Several trials have been performed by Meyer *et al.* (2003), Engelbrecht and Cloete (2016) and Engelbrecht *et al.* (2016) to investigate and quantify alternative factors such as on-farm related skin damage in an effort to improve ostrich leather quality. These authors found that cuts, scratches, kick marks and toenail-related injuries that occur during the grow-out phase of an ostrich resulted in permanent scars, leading to the downgrading of the skin and lower leather quality. Swart *et al.* (1984) stated that the harvesting of body feathers improved nodule development, but this practice exposes the skin to sunburn, which results in permanent scars and degrading of the ostrich leather. Ostrich quill mite and feather louse are parasites that are also responsible for lowering skin and leather quality (Cooper, 2005).

With regards to the meat composition of the birds that consumed the sweet lupin diet, mean moisture, crude protein and ash contents of 763.8 ± 1.07, 206.9 ± 1.00 and 11.7 ± 0.27 g/kg respectively were observed across all the treatments. Furthermore, no significant differences were observed for these chemical components for the different treatment groups. However, the average value for the intra-muscular fat content was 25.6 ± 1.37 g/kg and a significant difference was observed between the different dietary treatment groups. The intra-muscular fat content of the meat of the birds fed the 50LD (28.6 ± 1.24 g/kg) was the highest relative to the intra-muscular fat content of the birds receiving the 0LD (23.5 ± 1.24 g/kg) and 100LD (23.3 ± 1.24 g/kg).

According to Poławska *et al.* (2011), the most important factor influencing the fat content of the meat is the type of feed the birds receive. Horbańczuk and Sales (1998) stated that the tissue fatty acid composition of monogastric animals can easily be altered by feeding, particularly the level and type of energy source included in the diet. Hoffman (2005) reported that the chemical composition of ostrich meat is influenced by the nutrition of the bird, specifically with regards to the energy:protein ratio. It is known that the energy source can influence the fatty acid profile of ostrich meat, which is particularly visible in the various subcutaneous fat pads (Hoffman, 2005). The energy and protein contents of the different dietary treatments were within the boundaries for optimum nutrition, as predicted by the mathematical optimisation model developed by Gous and

Brand (2008). Further analysis should therefore be conducted to investigate the fatty acid profile of meat from ostriches fed diets containing sweet lupins.

The oil content of sweet lupins is 1.4% higher than that of soybean oilcake meal (Ewing, 1997; Feedipedia, 2016). For the starter, grower and finisher phases, the total average fat content of the control diets was 34.6 g/kg, compared to a total average fat content of 46.6 g/kg for the other diets. In other words, the total average fat content of the experimental diets containing sweet lupins was 34.4% higher than that of the control diets, which were formulated using soybean oilcake meal as the sole protein source. Although the diets were formulated to be iso-nutritious in terms of metabolisable energy, protein and amino acids, the intra-muscular fat content of the birds fed the intermediate diet was significantly higher than that of the birds fed the 0LD (control) and 100LD (maximum lupin inclusion level). This is contrary to the expectation that the birds fed the 100LD would have had the highest intra-muscular fat content. This finding is however difficult to explain, as Carstens (2013) and Viviers (2015) found no significant differences in the intra-muscular fat content of the meat of ostriches fed diets with different protein and energy contents.

Overall the findings for the chemical composition of the meat are consistent and supported by the findings of several previous studies. The results of these studies are summarised in Table 4.8.

Table 4.8. *The chemical composition (g/kg) of ostrich meat as found in various studies*

Chemical component (g/kg)	Sweet lupin inclusion trial	Sales and Oliver-Lyons (1996)	Sales and Hayes (1996)	Hoffman *et al*. (2005)	Majewska *et al*. (2009)	Dalle Zotte *et al*. (2013)
Moisture	763.8	777.0	762.7	766.6	767.0	750.2
Crude protein	206.9	206.0	211.2	216.6	213.0	212.0
Crude fat	25.6	2.60	6.50	20.5	9.00	30.9
Ash	11.7	11.6	10.7	12.2	11.0	10.7

4.5. CONCLUSION

Overall the results from this study are similar to the results from previously performed studies. These findings assure producers that the sweet lupin inclusion levels evaluated in this study will yield similar quality returns as for the standard commercial slaughter ostrich diets with soybean oilcake meal as the primary protein source. Currently the price of lupins is 56% of the price of soybean oilcake meal, making it a worthy economic alternative. Results from this study will contribute to the currently limited knowledge of the nutritional value of sweet lupins as a raw material in ostrich rations. The knowledge gained from this study can be used to formulate diets for ostriches more accurately and consequently improve the economical production of ostrich meat,

leather and feathers. In addition it will aid in developing the market for locally produced sweet lupins in South Africa, which will eventually benefit both the local grain industry as well as the ostrich industry.

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CHAPTER 5

FEED PREFERENCE OF GROWER OSTRICHES CONSUMING DIETS DIFFERING IN *LUPINUS ANGUSTIFOLIUS* INCLUSION LEVELS

Abstract

1. Feed costs make up the largest proportion (*ca*. 75%) of the input costs of slaughter birds in an intensive ostrich production unit. Alternative, cheaper feedstuffs, such as lupins (sweet and bitter cultivars), were therefore evaluated to determine to what degree lupins can be included in ostrich rations without affecting feed preference and intake.

2. Sixty South African Black ostriches were randomly divided into ten paddocks of six birds per paddock. Three trials with five different experimental diets per trial were conducted to investigate the diet preference of grower ostriches in a free-choice system. Feed and water was supplied *ad libitum*.

3. The position of the diets in the successive paddocks was varied by rotating the five feed troughs in a clockwise direction, but within each paddock the position of each feeder and diet stayed the same throughout the three trials.

4. In the first two trials sweet (trial 1) or bitter (trial 2) lupins replaced soybean oilcake meal to have 0%, 7.5%, 15%, 22.5% and 30% lupin inclusion levels in the diet. In trial 3 the soybean oilcake meal was replaced with either sweet or bitter lupins to have dietary inclusion levels as follows: 0% lupins, 15% sweet, 15% bitter, 30% sweet or 30% bitter. The daily intake per group for each diet was monitored over a period of five days each. The average initial body weight of the birds was 73.6 ± 0.5 kg.

5. No interaction was found between day and diet for the three trials and dry matter intake (DMI) did not differ between the five treatments for any of the three trials. In the second trial the birds tended (P = 0.11) to show a preference for the 7.5% bitter lupin inclusion level and discriminated against the 15% and 30% bitter lupin inclusion levels. Regression analysis of DMI on lupin inclusion rate revealed no significant trends.

6. In conclusion, the study revealed that soybean oilcake meal can be replaced in the diets of grower ostriches by sweet lupin inclusion levels up to 30%, without any significant detrimental effect on diet preference and feed intake.

5.1. INTRODUCTION

Feed costs make the largest proportion (*ca*. 75%) of the input costs of slaughter birds in an intensive ostrich production unit (Brand *et al.*, 2000; Brand and Gous, 2006; Jordaan *et al*., 2008). Volatile feed prices, seasonal droughts, exchange rates and market trends (consumer preference and economic cycles) have a large effect on the profitability of ostrich production. It is thus of cardinal importance to optimise the aspects of the ostrich production unit which can be controlled by the producer, such as nutrition (Carstens, 2013). Least-cost diet formulations and the use of alternative protein sources are two ways of decreasing feeding costs; however it is important that this does not have a detrimental effect on the quality of the end products.

Protein sources are becoming scarcer and more expensive, especially for use in animal feeds (Brand *et al*., 2004a). Depending on the feeding phase (pre-starter, starter, grower or finisher), protein composes up to 22.5% of a balanced diet (Brand and Gous, 2006). Ostrich producers are therefore looking for alternative, locally produced protein sources that are less expensive but still deliver acceptable production yields. Lupins have been identified as one possibility. They have already been successfully included in the diets of both monogastric (Brand *et al*., 1995) and ruminant animals (Brand *et al*., 1992; Brand *et al*., 1997), and in some cases have effectively replaced previously-used protein sources such as soybean oilcake meal. However, it is important to note that due to the presence of anti-nutritional factors such as alkaloids, lupins can only be included at certain levels for efficient utilisation and to prevent undesirable effects (Brand and Brandt, 2000). This problem is to some extent eliminated by the presence of sweet (low in alkaloids, < 0.1%) and bitter (alkaloid-rich, 0.1 – 4.0%) varieties within the species. To reduce the risk of toxicity an alkaloid level of less than 0.6 g/kg is deemed suitable for animal feeds (McDonald *et al*., 2011). The low alkaloid content of the sweet lupin varieties makes their use in diets with prolonged intakes of little concern regarding toxicity for the animal.

According to Ferguson *et al.* (2002), under certain circumstances commercial farm animals can make a rational choice between feeds according to their nutritional needs. Brand *et al.* (2004b) provided five types of lupins to young ostriches in a free-choice system and noted that in addition to smell and taste, colour and previous exposure to a certain type of feed may also have an influence on feed preference. It has been found in industry that young growing and finisher birds in feedlots refuse to eat feed when the composition or physical characteristics are suddenly changed. This phenomenon is generally observed when feed with a relatively green colour is changed to a feed that is less green in colour. Birds therefore need to be gradually exposed to a new feed to avoid a decrease in intake and consequently a drop in production (Brand, 2008).

Milton *et al*. (1994) found during a field study on food selection by ostriches in Southern Africa that ostriches did not feed on toxic plants. They assumed that the birds identified these

species by sight but also suggested that taste and smell may also play a role in determining the palatability of the feed, as while the ostriches were foraging they occasionally dropped plucked plant material. It therefore seems that when ostriches check the quality of a feed they conduct a preliminary visual inspection as well as utilising taste and smell. Chemoreceptive events in the mouth and olfactory epithelium are responsible for the final recognition and selection of the feed. These structures trigger the emotional experience of acute pleasure or displeasure (Kruger, 2007).

The aim of this study was therefore to determine the feed intake of ostriches when fed diets containing different levels of lupins (sweet and bitter cultivars), to determine to what degree lupins can be included in ostrich rations without affecting feed preference and intake.

5.2. MATERIALS AND METHODS

5.2.1. Experimental location and design

The trial was conducted in July 2015 at the Oudtshoorn Research Farm in the Little Klein Karoo region of South Africa (situated at longitude 22°15' E and latitude 33°37' S at an altitude of 300 m above sea level). The experimental design included 60 South African Black ostriches of 43 weeks of age that were randomly divided into 10 paddocks with six birds per paddock. The average initial body weight of the birds was 73.6 ± 0.5 kg. Ethical clearance (R14/108) for this study was granted by the Elsenburg ethical committee.

5.2.2. Experimental procedure and dietary treatment

Three trials with five experimental diets per trial were conducted to investigate the diet preference of grower ostriches in a free-choice system. In Trial 1 and 2, soybean oilcake meal was replaced by sweet (Trial 1) or bitter (Trial 2) lupins in a step-wise manner to produce five experimental diets with lupin inclusion levels of 0%, 7.5%, 15%, 22.5% and 30%. Trial 3 differed from Trial 1 and 2 in that soybean oilcake meal was replaced by alternatively sweet or bitter lupins to provide diets with 0% lupins, 15% sweet, 15% bitter, 30% sweet and 30% bitter lupin inclusion levels. The positions of the feeders containing each diet in the successive paddocks were altered by rotating the five feed troughs in a clockwise direction, but within each paddock the specific position of each feeder and diet stayed the same throughout the three trials. The feed troughs were spread more or less evenly within each paddock and provided sufficient feeding space per bird. The approximate dimensions of the feed troughs were 46 cm x 23 cm x 20 cm and of the paddocks, 32 m x 30 m. Both feed and water were provided *ad libitum*, with the dimensions of the water buckets being 29 cm x 20 cm x 15 cm.

The daily intake of each diet per group was monitored over a period of five days for each trial. This was done by weighing back the refusals of the day and subtracting it from the amount of feed offered during the day. The feed in the feed troughs was mixed twice daily (early in the morning and at midday) by hand to stimulate feed intake. The recording of feed provided and feed refused occurred at the same time every day. Despite inclement weather conditions on certain days of the trial periods, data capturing was completed without disruptions.

5.2.3. Dietary formulation and chemical components

The treatment diets were formulated using Mixit2+ software (Agricultural Software Consultants Inc., San Diego, USA). The raw material composition of the sweet and bitter lupins and soybean oilcake meal used in the diet formulations of this study can be found in Table 5.1. The raw material samples were grounded using a RetschTM ZM200 sample mill (Haan, Germany) with a 1.5 mm screen to create a meal with a consistent particle size. Thereafter the raw materials were analysed using the methods of the Association of Official Analytical Chemists (AOAC, 2002) for dry matter (DM) (method 934.01), ash (method 942.05), crude protein (CP) (method 976.05), crude fibre (CF) (method 962.09), ether extract (EE) (method 920.39), acid detergent fibre (ADF) (Goering and van Soest, 1970), neutral detergent fibre (NDF) (Robertson and van Soest, 1981). The calcium (Ca) and phosphorous (P) values were analysed using method 6.1.1 (Dry Ashing) of the Agri Laboratory Association of Southern Africa guidelines (ALASA) (ALASA, 1998).

Table 5.1. *Raw material composition of the sweet and bitter lupins and soybean oilcake meal used in the diet formulations of this study*

Nutrient component (g/kg)	Sweet lupins	Bitter lupins	Soybean oilcake meal
Dry matter	902.5	898.7	910.8
Ash	29.5	26.2	62.5
Crude protein	309.4	313.8	463.1
Crude fibre	154.0	156.3	32.0
Ether extract	48.9	42.2	10.3
Neutral detergent fibre	244.8	231.9	81.9
Acid detergent fibre	196.9	189.4	44.3
Calcium	2.60	2.60	2.90
Phosphorous	4.90	4.50	8.30

The formulations of the diets are presented in Tables 5.2 – 5.4. The experimental diets were mixed, milled and pelleted at the Kromme Rhee Research Farm (situated 18°50' E, 33°37' S with an altitude of 177 m above sea level) and transported to the Oudtshoorn Research Farm. Tables 5.2 – 5.4 also provide the nutritional compositions of the experimental diets. These values were determined for samples randomly collected during the feed mixing and pelleting process. The samples were ground using a RetschTM ZM200 sample mill (Haan, Germany) with a 1.5 mm screen to create a meal with a consistent particle size. Thereafter, all samples of the same experimental diet were pooled and analysed for dry matter (DM), crude protein (CP), ether extract (EE), ash, crude fibre (CF), acid detergent fibre (ADF), neutral detergent fibre (NDF) and *in vitro* organic matter digestibility (IVOMD) using a Bran + Luebbe InfrAlyzer 500 near infrared reflectance spectrometer (IA-500) (NIRS). The samples (*ca.* 6.0 g) were individually presented in closed cups and scanned in the reflectance mode at between 1100 – 2500 nm in the near-infrared region with 2 nm intervals, acquiring 701 data points for each sample. The spectroscopic measurements were interpreted using Bran + Leubbe SESAME Version 2.00 software (Bran + Luebbe GmbH, Norderstedt, Germany). The ME (MJ/kg feed) was calculated using the following equation: ME = 0.015 x IVOMD (g/kg DM) (Van der Honing and Alderman, 1988). The IVOMD was determined by an adaptation of the method of the two-stage rumen fluid-pepsin technique described by Tilley and Terry (1963). It involves firstly a 48 hour fermentation by rumen micro-organisms in a buffer solution, followed by a 48 hour pepsin-hydrochloric acid digestion. The residue represents the indigestible part of the sample.

5.2.4. Alkaloid content of the feed

The total alkaloid contents of the finely-ground pooled feed samples containing either the sweet (Eureka) or bitter (SSL 10) *Lupinus angustifolius* cultivars were determined as described by Boschin *et al*. (2008), with minor modifications. The sample preparation method was modified by extracting the total alkaloid content directly using a 50:50 methylene dichloride:methanol mixture (MDC:MeOH). GC-MS with a 30 m x 0.25 mm, internal diameter 0.25 μm, AT-Wax capillary column was then used to analyse the total alkaloid content. The temperature program was as follows: 150°C for 5 minutes, increased by 5°C per minute up to 300°C then maintained at 300°C for 15 minutes. Analyses were performed in split mode with a split ratio of 1:25. The injection volume was 1 μL, injection temperature 250°C, interface temperature 300°C and the acquisition was from *m*/*z* 50 to 450. The source operated in EI mode at eV. The total alkaloids were identified using Mass library (Agilent) and the detection limit for quantifying the total alkaloids was 100 g/ml. However, no alkaloids were found in the respective feed samples at this detection limit. Therefore, the sweet and bitter lupin cultivars used in this study was the same cultivars (sweet *L. angustifolius*

and bitter *L. angustifolius*) used by Brand and Smith (2016). The spectrophotometry method described by Von Baer *et al*. (1978) was used to determine the total alkaloid content of these cultivars in the study by Brand and Smith (2016). This method is a quantitative determination of total alkaloids with bromocresol purple at 405 nm. The total alkaloid content of the sweet and bitter lupin cultivars in the study by Brand and Smith (2016) was 49.1 mg/kg and 15 204.5 mg/kg respectively. These values were used to calculate the estimated amount of total alkaloids of the five dietary treatments of this study for each of the three trials (Tables 5.2 – 5.4).

Table 5.2. *The formulation and nutritional composition (as fed basis) of five treatment diets containing different sweet lupin inclusion levels fed to grower phase slaughter ostriches (Trial 1)*

Raw materials (kg/ton)	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (7.5%)	3 (15%)	4 (22.5%)	5 (30%)
Maize meal	590.6	544.9	499.2	453.5	407.8
Soybean oilcake meal	149.3	111.0	74.7	37.3	0.00
Sweet lupins	0.00	76.5	152.9	226.5	300.0
Lucerne meal	186.4	193.5	200.5	210.6	220.7
Molasses powder	25.0	25.0	25.0	25.0	25.0
Monocalcium phosphate	17.4	17.2	16.6	15.9	15.2
Limestone	14.5	14.8	15.0	15.3	15.5
Salt	10.0	10.0	10.0	10.0	10.0
Synthetic lysine	0.87	0.76	0.65	0.53	0.42
Synthetic methionine	0.41	0.43	0.45	0.46	0.48
Mineral and vitamin premix*	5.00	5.00	5.00	5.00	5.00
Nutrient component					
DM[1] (g/kg)	893.9	911.2	900.1	911.8	901.5
ME MJ/kg feed[2]	13.4	13.7	13.5	13.7	13.5
IVOMD[3] (g/kg)	851.2	838.1	841.3	825.5	827.2
CP[4] (g/kg)	160.3	163.5	169.9	159.9	175.8
Ash (g/kg)	90.3	99.3	99.5	111.1	100.8
EE[5] (g/kg)	22.8	28.0	26.8	28.2	33.1
CF[6] (g/kg)	67.1	89.2	74.7	88.9	88.1
ADF[7] (g/kg)	97.6	124.3	108.1	130.5	119.7
NDF[8] (g/kg)	166.4	206.5	173.0	204.2	188.0
Total alkaloid content (ppm)	0.00	3.68	7.37	11.0	14.7

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for grower ostriches

[1]Dry matter

[2]Metabolisable energy

[3]*In vitro* organic matter digestibility

[4]Crude protein

[5]Ether extract

[6]Crude fibre

[7]Acid detergent fibre

[8]Neutral detergent fibre

Table 5.3. *The formulation and nutritional composition (as fed basis) of five treatment diets containing different bitter lupin inclusion levels fed to grower phase slaughter ostriches (Trial 2)*

Raw materials (kg/ton)	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (7.5%)	3 (15%)	4 (22.5%)	5 (30%)
Maize meal	590.6	544.9	499.2	453.5	407.8
Soybean oilcake meal	149.3	112.0	74.7	37.3	0.00
Bitter lupins	0.00	76.5	152.9	226.5	300.0
Lucerne meal	186.4	193.5	200.5	210.6	220.7
Molasses powder	25.0	25.0	25.0	25.0	25.0
Monocalcium phosphate	17.4	17.2	16.6	15.9	15.2
Limestone	14.5	14.8	15.0	15.3	15.5
Salt	10.0	10.0	10.0	10.0	10.0
Synthetic lysine	0.87	0.76	0.65	0.53	0.42
Synthetic methionine	0.41	0.43	0.45	0.46	0.48
Vitamin and vitamin premix*	5.00	5.00	5.00	5.00	5.00
Nutrient component					
DM[1] (g/kg)	893.9	902.6	898.0	908.0	902.0
ME MJ/kg feed[2]	12.8	12.6	12.6	12.2	12.3
IVOMD[3] (g/kg)	851.2	839.0	839.7	810.5	822.2
CP[4] (g/kg)	160.3	170.2	172.1	168.3	185.3
Ash (g/kg)	90.3	94.5	102.9	98.3	103.8
EE[5] (g/kg)	22.8	26.9	28.6	31.0	29.5
CF[6] (g/kg)	67.1	84.4	79.1	102.9	96.1
ADF[7] (g/kg)	97.6	115.7	112.4	134.8	126.6
NDF[8] (g/kg)	166.4	192.5	186.7	210.0	199.4
Total alkaloid content (ppm)	0.00	1 140.3	2 280.7	3 421.0	4 561.4

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for grower ostriches

[1]Dry matter

[2]Metabolisable energy

[3]*In vitro* organic matter digestibility

[4]Crude protein

[5]Ether extract

[6]Crude fibre

[7]Acid detergent fibre

[8]Neutral detergent fibre

Table 5.4. *Formulation and nutritional composition (as fed basis) of five treatment diets containing sweet or bitter lupins at different inclusion levels fed to grower phase slaughter ostriches (Trial 3)*

Raw materials (kg/ton)	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (15% Sweet)	3 (15% Bitter)	4 (30% Sweet)	5 (30%Bitter)
Maize meal	590.6	544.9	544.9	407.8	407.8
Soybean oilcake meal	149.3	112.0	112.0	0.00	0.00
Sweet lupins	0.00	76.5	0.00	300.0	0.00
Bitter lupins	0.00	0.00	76.5	0.00	300.0
Lucerne meal	186.4	193.5	193.5	220.7	220.7
Molasses powder	25.0	25.0	25.0	25.0	25.0
Monocalcium phosphate	17.4	17.2	17.2	15.2	15.2
Limestone	14.5	14.8	14.8	15.5	15.5
Salt	10.0	10.0	10.0	10.0	10.0
Synthetic lysine	0.87	0.76	0.76	0.42	0.42
Synthetic methionine	0.41	0.43	0.43	0.48	0.48
Mineral and vitamin premix*	5.00	5.00	5.00	5.00	5.00
Nutrient component					
DM[1] (g/kg)	893.9	900.1	898.0	901.5	902.0
ME MJ/kg feed[2]	12.8	12.6	12.6	12.4	12.3
IVOMD[3] (g/kg)	851.2	841.3	839.7	827.2	822.2
CP[4] (g/kg)	160.3	169.9	172.1	175.8	185.3
Ash (g/kg)	90.3	99.5	102.9	100.8	103.8
EE[5] (g/kg)	22.8	26.8	28.6	33.1	29.5
CF[6] (g/kg)	67.1	74.7	79.1	88.1	96.1
ADF[7] (g/kg)	97.6	108.1	112.4	119.7	126.6
NDF[8] (g/kg)	166.4	173.0	186.7	188.0	199.4
Total alkaloid content (ppm)	0.00	7.37	2 280.7	14.7	4 561.4

*Refer to APPENDIX 1 for the composition of the vitamin and mineral premix for grower ostriches

[1]Dry matter, [2]Metabolisable energy, [3]*In vitro* organic matter digestibility, [4]Crude protein, [5]Ether extract, [6]Crude fibre, [7]Acid detergent fibre, [8]Neutral detergent fibre

5.2.5. Mineral content of the feed

The mineral compositions and amino acid profiles of the diets are presented in Tables 5.5 – 5.7. To determine the mineral content of the pooled reference samples the finely ground (passed through a 1.5 mm sieve) feed samples were analysed using method 6.1.1 (Dry Ashing) of the Agri Laboratory Association of Southern Africa guidelines (ALASA) (ALASA, 1998). A 1 – 3 g sample of each diet was weighed and placed in a porcelain crucible. The crucibles were placed in a muffle furnace and left to ash overnight at 460 – 480°C. Once the samples had cooled down, 5 ml of 1:1 hydrogen chloride (HCl) were added to each crucible to dissolve the samples. The crucibles were then placed in an oven for 30 minutes at 60°C for evaporation to take place. The samples were left to cool and then filled to a total volume of 40 ml with deionized water and mixed thoroughly before being filtered into an amber bottle. The mineral concentrations was measured using a Thermo Electron iCAP 6000 Series Inductively Coupled Plasma (ICP) Spectrophotometer (Thermo Electron Corporation, Milan, Italy) fitted with a vertical quartz torch and Cetac ASX-520 autosampler. Concentrations were determined using Merck Titrisol standards with concentrations of 1000 ppm (Merck, Darmstadt, Germany) and calculated using iTEVA Analyst software.

5.2.6. Amino acid profiles of the feed

The amino acid profiles were determined using the method described by Grace Davison (2008), through hydrolysis of the samples in hydrochloric acid and high pressure liquid chromatography (HPLC). In a hydrolysis tube, 6 ml of 6 N HCl and 15% Phenol solution was added to 0.1 g of feed sample. Nitrogen was then added and the samples were placed under vacuum. This was done by flushing the hydrolysis tube with the nitrogen to remove oxygen and create an anaerobic environment. The sealed hydrolysis tubes were then placed in an oven for 24 hours at 110°C to allow complete protein hydrolysis. The samples were left to cool and then filtered using a hydrophylic polyvinylidene difluoride syringe filter (PVDF - 0.45 µm, 33 mm) before being transferred into 1.5 ml Eppendorf tubes. Amino acids were derivatised with *o*-phthalaldehyde and 3-mercaptopropionic acid in borate buffer (Agilent Technologies, Waldbronn, Germany). Reverse-phase Dionex HPLC (Dionex Corporation, California, USA) was used to separate the amino acids on a 3.9 x 150 mm C18 Nova-Pak column (Waters, Ireland) at a 1.1 ml/minute flow rate. L-Amino acid standards (2.5 μmol/ml in 0.1 N HCl) (Thermo Scientific, Illinois, USA) were used to identify the amino acids.

Table 5.5. The *mineral and amino acid composition (as fed basis) of feeds containing five inclusion levels of sweet lupins fed to grower phase slaughter ostriches (Trial 1)*

	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (7.5%)	3 (15%)	4 (22.5%)	5 (30%)
Minerals					
Calcium (g/kg)	12.2	15.2	14.9	12.2	14.4
Phosphorous (g/kg)	6.00	7.60	7.40	6.40	7.25
Magnesium (g/kg)	2.30	2.60	2.50	2.50	2.50
Sodium (g/kg)	3.88	4.11	5.01	3.56	4.86
Manganese (mg/kg)	210.9	279.8	257.8	345.6	273.6
Copper (mg/kg)	14.3	16.8	16.8	20.3	19.3
Iron (mg/kg)	333.5	332.4	393.4	357.8	268.6
Zinc (mg/kg)	119.0	151.4	137.7	170.2	147.8
Amino acids (g/kg)					
Lysine	9.80	14.0	17.2	14.7	24.6
Methionine	0.30	0.60	0.20	0.10	0.10
Arginine	6.50	8.20	10.0	8.60	13.5
Threonine	4.90	5.50	6.10	5.20	7.30
Tyrosine	5.10	5.80	6.50	5.70	7.90
Aspartic acid	13.0	14.9	16.1	13.6	19.2
Glutamic acid	19.5	23.3	26.4	22.8	33.3
Serine	6.10	7.00	7.90	6.70	9.70
Histidine	2.30	2.60	2.70	2.20	3.10
Glycine	5.30	6.10	6.80	5.80	8.30
Alanine	6.10	6.80	7.40	6.50	8.70
Valine	6.30	7.10	7.60	6.50	8.90
Phenylalanine	6.60	7.40	7.90	6.80	9.20
Isoleucine	5.20	6.00	6.50	5.50	7.80
Leucine	11.2	12.6	13.7	12.1	16.2

Table 5.6. *The mineral and amino acid composition (as fed basis) of feeds containing five inclusion levels of bitter lupins fed to grower phase slaughter ostriches (trial 2)*

	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (7.5%)	3 (15%)	4 (22.5%)	5 (30%)
Minerals					
Calcium (g/kg)	12.2	15.2	14.9	12.2	14.4
Phosphorous (g/kg)	6.00	7.60	7.40	6.40	7.25
Magnesium (g/kg)	2.30	2.60	2.50	2.50	2.50
Sodium (g/kg)	3.88	4.11	5.01	3.56	4.86
Manganese (mg/kg)	210.9	262.8	262.9	241.2	321.2
Copper (mg/kg)	14.3	15.8	18.1	15.8	17.2
Iron (mg/kg)	333.5	322.2	385.2	371.8	304.3
Zinc (mg/kg)	119.0	137.7	145.6	120.6	161.8
Amino acids (g/kg)					
Lysine	9.80	13.1	16.4	13.5	23.0
Methionine	0.30	0.24	0.18	0.20	0.06
Arginine	6.50	8.10	9.70	8.78	12.9
Threonine	4.90	5.26	5.63	4.72	6.35
Tyrosine	5.10	5.61	6.13	5.20	7.15
Aspartic acid	13.0	14.2	15.3	13.3	17.6
Glutamic acid	19.5	22.7	25.8	21.9	32.1
Serine	6.10	6.82	7.54	6.36	8.98
Histidine	2.30	2.41	2.51	1.99	2.72
Glycine	5.30	5.87	6.44	5.46	7.59
Alanine	6.10	6.38	6.66	5.56	7.21
Valine	6.30	6.65	7.01	6.08	7.72
Phenylalanine	6.60	7.15	7.71	7.40	8.82
Isoleucine	5.20	5.66	6.11	5.22	7.02
Leucine	11.2	12.0	12.7	10.5	14.2

Table 5.7. *The mineral and amino acid composition (as fed basis) of feeds containing five inclusion levels of either sweet or bitter lupins fed to grower phase slaughter ostriches (trial 3)*

	Diet number and percentage lupin inclusion level				
	1 (0%)	2 (15% Sweet)	3 (15% Bitter)	4 (30% Sweet)	5 (30% Bitter)
Minerals					
Calcium (g/kg)	12.2	14.3	14.9	12.9	14.4
Phosphorous (g/kg)	6.60	7.20	7.40	6.90	7.25
Magnesium (g/kg)	2.30	2.50	2.50	2.40	2.50
Sodium (g/kg)	3.88	4.89	5.01	5.31	4.86
Manganese (mg/kg)	210.9	257.8	262.9	273.6	321.2
Copper (mg/kg)	14.3	16.8	18.1	19.3	17.2
Iron (mg/kg)	333.5	393.4	385.2	268.6	304.3
Zinc (mg/kg)	119.0	137.7	145.6	147.8	161.8
Amino acids (g/kg)					
Lysine	9.80	17.2	16.4	24.6	23.0
Methionine	0.30	0.20	0.18	0.10	0.06
Arginine	6.50	10.0	9.70	13.5	12.9
Threonine	4.90	6.10	5.63	7.30	6.35
Tyrosine	5.10	6.50	6.13	7.90	7.15
Aspartic acid	13.0	16.1	15.3	19.2	17.6
Glutamic acid	19.5	26.4	25.8	33.3	32.1
Serine	6.10	7.90	7.54	9.70	8.98
Histidine	2.30	2.70	2.51	3.10	2.72
Glycine	5.30	6.80	6.44	8.30	7.59
Alanine	6.10	7.40	6.66	8.70	7.21
Valine	6.30	7.60	7.01	8.90	7.72
Phenylalanine	6.60	7.90	7.71	9.20	8.82
Isoleucine	5.20	6.50	6.11	7.80	7.02
Leucine	11.2	13.7	12.7	16.2	14.2

5.2.7. Colour of the feed

The CIE Lab-System describes colour according to three surface colour attributes namely L* (lightness), a* (redness) and b* (yellowness). The L* coordinate represents the lightness (reflection) of the sample, where 0 = black and 100 = white. The a* coordinate signifies the red/green spectrum, where a positive value indicates the degree of redness and a negative value indicates that green pigments are being detected. The b* coordinate characterises the yellow/blue range, with a positive value indicating the degree of yellowness and a negative value indicating the degree of blueness (BYK-Gardner GmbH). The surface colour of the finely ground feed samples was measured using a colour-guide 45°/0° colorimeter with an aperture size of 20 mm and an illuminant/observer ratio of D65/10° (Catalogue number 6805, BYK-Gardner GmbH, Geretsried, Germany). Calibration of the colorimeter was done using the standards provided (BYK-Gardner). The finely ground sample was spread evenly in a petri dish and five repeats were taken per measurement per sample. The results are presented in Table 5.8.

5.2.8. Statistical analysis

Statistical analysis was performed using Statgraphics Centurion (Version 15; Statpoint, Inc., Virginia, USA), SAS Enterprise Guide (Version 9.2; SAS Institute Inc., Cary, USA) and Microsoft Office Excel 2010 (Version 14.0, Microsoft Corporation by Impressa Systems, Santa Rosa, California). Descriptive statistics were performed on the respective CIE Lab-System colour attributes (L*, a* and b*) per diet for each trial to determine whether the diets in the respective trials were similar in colour. A multifactor analysis of variance (ANOVA) was done for all three trials separately to determine which of the two main effects, day and diet, had a statistically significant effect on the mean dry matter intake (DMI) per bird per treatment diet. The multifactor ANOVA was also used to test whether there was a significant interaction between the two main effects. A one-way ANOVA was conducted to evaluate the mean DMI as well as the %DMI per bird per day by diet. A regression analysis of the mean DMI per bird was done per treatment diet over the different lupin inclusion levels (%) for Trial 1 (sweet lupins) and 2 (bitter lupins). Statistical differences were declared at $P < 0.05$.

5.3. RESULTS

Regarding the colour attributes in Trial 1, the L* value of diet 2 differed ($P < 0.05$) from that of diets 1, 3 and 4. No differences were observed for the a* attribute between the respective diets. The b* attribute of diet 3 differed ($P < 0.05$) from diets 1, 4 and 5, while diet 5 also differed ($P < 0.05$) from diets 1, 2 and 3 (Table 5.8). In Trial 2, the L* attribute of diet 3 differed ($P < 0.05$) from that of diet 5. The a* attribute of diets 1 and 2 differed ($P < 0.05$) from diet 3, while the b* attribute of diet 5 differed ($P < 0.05$) from the remaining four diets (Table 5.8). In Trial 3, the L* attribute of diet 5 differed ($P < 0.05$) from diets 2 and 3, while the a* attribute of diet 3 differed ($P < 0.05$) from diets 1 and 2. The b* attribute of diet 1 and 3 differed ($P < 0.05$) from diets 2, 4 and 5, while diets 4 and 5 differed ($P < 0.05$) from diets 1, 2 and 3. Diet 2 differed ($P < 0.05$) from all four other diets (Table 5.8). However, while statistically significant differences were observed between the diets for the colour attributes, these differences were small.

Table 5.8. *Descriptive statistics of the CIE Lab-System colour attributes (L*, a* and b*) for diets with varying sweet and bitter lupin inclusion levels*

Descriptive Statistics and trial	CIE Lab-System colour attributes per diet and percentage lupin inclusion level														
	1 L*	2 L*	3 L*	4 L*	5 L*	1 a*	2 a*	3 a*	4 a*	5 a*	1 b*	2 b*	3 b*	4 b*	5 b*
Trial 1: Sweet lupins	0%	7.5%	15%	22.5%	30%	0%	7.5%	15%	22.5%	30%	0%	7.5%	15%	22.5%	30%
Mean	57.0	56.4	55.5	60.4	59.4	3.1	3.3	4.1	3.6	3.7	19.7	20.7	19.9	23.5	22.8
Standard error of the mean	0.90	0.50	0.54	1.14	0.26	0.20	0.09	0.09	0.21	0.11	0.14	0.04	0.17	0.45	0.29
Lower bound on mean (95%)	54.5	55.0	54.0	57.2	58.7	2.6	3.1	3.8	3.0	3.3	19.3	20.6	19.5	22.3	22.0
Upper bound on mean (95%)	59.5	57.8	57.0	63.6	60.1	3.7	3.6	4.3	4.2	4.0	20.1	20.9	20.4	24.8	23.6
Trial 2: Bitter lupins	0%	7.5%	15%	22.5%	30%	0%	7.5%	15%	22.5%	30%	0%	7.5%	15%	22.5%	30%
Mean	57.0	56.3	55.5	55.1	59.4	3.1	2.6	4.1	3.6	3.7	19.7	20.3	19.9	19.7	22.8
Standard error of the mean	0.90	1.32	0.54	1.29	0.26	0.20	0.30	0.09	0.29	0.11	0.14	0.34	0.17	0.70	0.29
Lower bound on mean (95%)	54.5	52.6	54.0	51.5	58.7	2.6	1.7	3.8	2.7	3.3	19.3	19.4	19.5	17.7	22.0
Upper bound on mean (95%)	59.5	59.9	57.0	58.6	60.1	3.7	3.4	4.3	4.4	4.0	20.1	21.3	20.4	21.6	23.6
Trial 3: Sweet and bitter lupins	0%	15% Sweet	15% Bitter	30% Sweet	30% Bitter	0%	15% Sweet	15% Bitter	30% Sweet	30% Bitter	0%	15% Sweet	15% Bitter	30% Sweet	30% Bitter
Mean	57.0	56.4	55.5	60.4	59.4	3.1	3.3	4.1	3.6	3.7	19.7	20.7	19.9	23.5	22.8
Standard error of the mean	0.90	0.50	0.54	1.14	0.26	0.20	0.09	0.09	0.21	0.11	0.14	0.04	0.17	0.45	0.29
Lower bound on mean (95%)	54.5	55.0	54.0	57.2	58.7	2.6	3.1	3.8	3.0	3.3	19.3	20.6	19.5	22.3	22.0
Upper bound on mean (95%)	59.5	57.8	57.0	63.6	60.1	3.7	3.6	4.3	4.2	4.0	20.1	20.9	20.4	24.8	23.6

Regarding feed intake, no interaction was found between the two main effects (day and diet) in any of the three trials (P = 0.45, 0.88 and 0.99 for trials 1, 2, and 3 respectively). The main effects were therefore investigated individually and one-way ANOVA's were conducted to evaluate the effect of diet on the mean DMI and %DMI per bird per day. The results for both mean DMI and %DMI indicated that feed intake did not differ between the five diets for any of the three trials (Table 5.9). There was, however, a tendency (P = 0.11) during the second trial for birds to show a preference for the 7.5% bitter lupin inclusion level and discriminate to some extent against the 15% and 30% inclusion levels (Table 5.9).

Table 5.9. *Least square means ± standard error (LSM ± SE) for the effect of sweet and bitter lupin inclusion levels on the mean DMI and %DMI of grower phase slaughter ostriches*

Treatment			Mean DMI/bird/day (g)	Percentage of DMI/bird/day (%)
Lupin variety	Diet	Lupin inclusion level (%)		
Sweet Trial 1	1	0	541.13 ± 35.43	18.11 ± 1.13
	2	7.5	646.20 ± 35.43	21.52 ± 1.13
	3	15	628.07 ± 35.43	20.92 ± 1.13
	4	22.5	583.70 ± 35.43	19.62 ± 1.13
	5	30	594.03 ± 35.43	19.83 ± 1.13
Bitter Trial 2	1	0	776.10^{ab} ± 78.27	21.30^{ab} ± 2.18
	2	7.5	890.60^{a} ± 78.27	24.46^{a} ± 2.18
	3	15	606.57^{b} ± 78.27	16.73^{b} ± 2.18
	4	22.5	695.53^{ab} ± 78.27	19.55^{ab} ± 2.18
	5	30	657.37^{b} ± 78.27	17.97^{b} ± 2.18
Sweet & Bitter Trial 3	1	0	893.00 ± 90.07	23.60 ± 2.34
	2	15 (Sweet)	628.23 ± 90.07	16.77 ± 2.34
	3	15 (Bitter)	672.00 ± 90.07	17.84 ± 2.34
	4	30 (Sweet)	736.97 ± 90.07	19.87 ± 2.34
	5	30 (Bitter)	808.90 ± 90.07	21.92 ± 2.34

^{a,b} Column means with different superscripts differ significantly ($P < 0.05$)

Regression analysis of DMI per bird per day on lupin inclusion level for trials 1 and 2 revealed no significant trend (Figure 5.1). A polynomial regression was fitted to both the trial 1 and 2 DMI data, but the quadratic function was non-significant in both cases (P = 0.47 and 0.62

respectively). The regression equation for the first trial accounted for 52.56% of the variance while the regression equation for the second trial accounted for 38.45%. These regression models therefore did not fit the data closely and do not describe the effect that lupin inclusion level has on DMI very accurately.

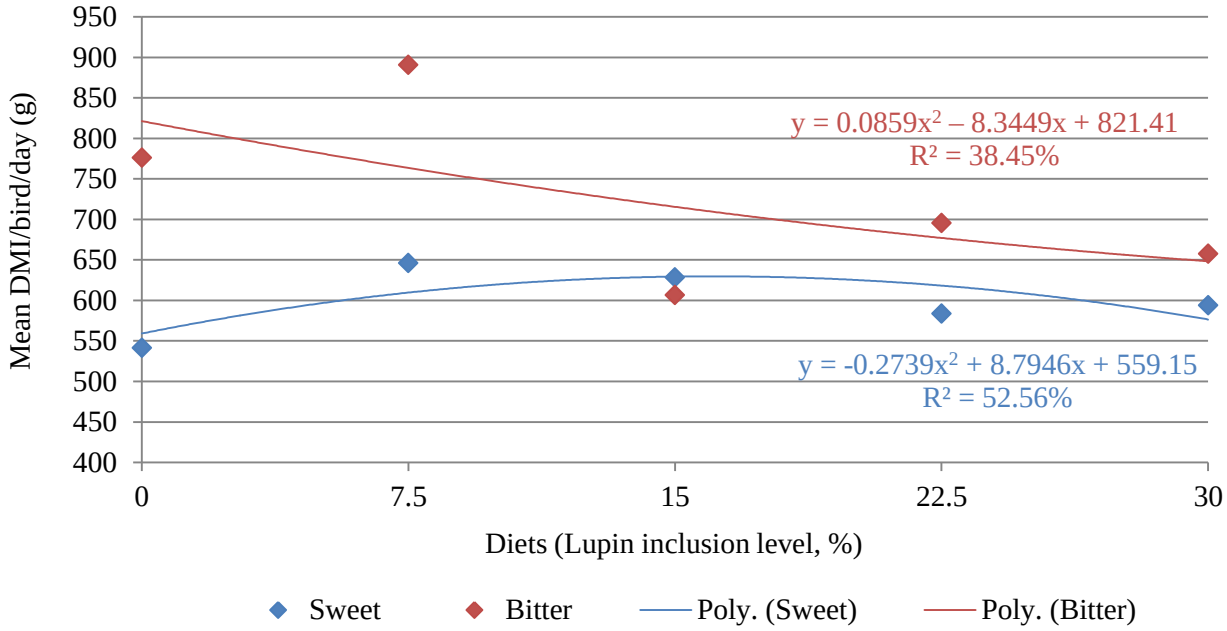

Figure 5.1. *Quadratic functions fitted to the DMI of grower phase slaughter ostriches fed diets containing different inclusion levels of sweet and bitter lupins*

5.4. DISCUSSION

The results of this study indicated that the feed preference and intake of ostriches is not influenced by the inclusion of sweet lupins in the diet up to 30%. This is unexpected as alkaloids are bitter tasting compounds, reducing the palatability of the feed (Smith, 2005). Previous studies on poultry and pigs found that lupin inclusion tended to reduce palatability, which result in poor acceptance and growth rates (Petterson and Fairbrother, 1996). The results found in Trial 1 of this study may be due to the low alkaloid content of the sweet lupin diets used. The prolonged consumption of the sweet lupin diets should therefore be of no concern regarding toxicity for the birds and can thus be regarded as safe and may therefore have prevented any effect on palatability from occurring.

Kruger *et al*. (2008) fed a conventional pre-starter mash diet (as formulated by Brand, 2005) artificially flavoured with four different commercially-produced non-toxic food flavourants (sweet, bitter, salty and sour) to ostrich chicks in a free-choice setup with no previous exposure to any of the feeds. There were five flat-bottomed feeders per pen in which the four flavoured diets and the control diet (untreated mash) were provided. It was concluded that the chicks preferred salty feed

(34.0%), then sweet (17.9%), control (17.1%), bitter (15.7%), and sour (15.4%). This could be attributed to the evolution of ostriches in deserts where the availability of good quality water is limited, as ostriches are able to utilise water with high salt levels due to their salt-excretory nasal glands (Kruger, 2007). It was not considered likely that the choice of salty food was directly related to flavour as Brand *et al.* (2008b) found that there were no conventional taste buds present in either two-month-old chicks or adult ostriches. If it is likely that ostriches can't taste, the tendency for the ostriches to prefer the 7.5% bitter lupin inclusion level and discriminate to some extend against the 15% and 30% inclusion levels (Trial 2; Table 5.9) could therefore reflect the small sample size used rather than any particular pattern of feed-choice. It is therefore possible that some other factor of the feed apart from its visual appearance, palatability thereof or the birds' previous experience may have played a role in producing this trend, because the bitter lupin diets had a much higher alkaloid content compared to the sweet lupin diets. Discrimination against the bitter lupin diets would thud almost be expected, but during Trial 3 both the mean DMI and %DMI indicated that feed intake did not differ between the five diets.

However, Tadjalli *et al*. (2008) studied the oropharyngeal cavity and its components (beak, hard palate, pharynx, tongue and the larynx) and found that the caudal third portion of the hard palate contains a semi circular darker area that is covered by many small delicate papillae. The other two thirds of the rostral part of the hard palate, that divides it into two regions, lack papillae. The ostrich also lack a transverse row of papillae caudal to the infundibular opening at the junction with the oesophagus. Furthermore, Gentle (1971) stated that chickens have a good sense of taste and Kare and Pick (1960) noted that when it comes to bitter-tasting substances in feed, a very high concentration is required over long periods before any reduction in feed intake is observed, despite the selection against bitter components when under free-choice conditions. The tendency observed during Trial 2 may therefore warrant further research, since the question of how important taste is in determining palatability in this species might rise.

Ferguson *et al*. (2002) noted that when young pigs were given a choice of diets, they instinctively avoided potentially harmful substances (toxins), anti-nutritional factors or unpalatable components in the feeds. Thereafter, they selected more nutritionally balanced feeds or feeds with more favourable amino acid profiles that would satisfy their requirements for growth and production. The iso-nitrogenous nature of the diets used in this study, the low alkaloid contents found in the sweet lupin diets, as well as the seemingly absence of taste in ostriches (could thus explain the lack of variation in DMI between the diets.

Evaluating lupins in the diets of pigs and poultry, it was found that the maximum lupin inclusion levels for pigs, as recommended by Australian researches, are as follows: 10-15% in starter diets, 20-25% in grower diets, 30-35% in finisher diets and 20% in dry and lactating sow

diets (Petterson and Fairbrother, 1996). In addition inclusion levels of up to 25% of low-alkaloid lupin-seed meal can be tolerated by broiler chickens without affecting growth unfavourably (Brenes *et al.*, 1993). Research has shown that a maximum inclusion level of 25-35% of either *L. angustifolius* or *L. albus* will not affect the laying performance of hens (Edwards and van Barneveld, 1998)., while in broiler chicken diets it should not exceed 10%.

An additional factor that may have been responsible for the no significant differences in feed preference and intake between the diets was their formulation to be iso-nutritious. Forbes and Shariatmadari (1994) state that when a single feed is provided, the intake is determined primarily by the energy content thereof. This is supported by Rose and Kyriazakis (1991), who reported that one of the major factors determining the diet selection of poultry and pigs are their nutrient requirements. Brand *et al*. (2012) consequently assumed that male and female South African Black ostriches would select feeds under free-choice feeding conditions according to their protein and energy requirements. In this study, the treatment diets were formulated to be iso-caloric (equal ME levels) and meet the requirements of the birds (Table 5.2 – 5.4). The diets did differ slightly in terms of their CP, fat and CF contents, but these differences were unlikely to have been great enough to have had a significant effect on diet selection and DMI (Table 5.2 – 5.4). This supports the results, which indicated that DMI and %DMI did not differ between the five treatments in any of the three trials. It also suggests that the diets provided in all three trials satisfied the nutrient requirements of the birds.

Although significant differences were observed in the CIE Lab-System values between the feeds they did not differ to any great extent under visual inspection (light yellowish-brown) (Table 5.8). The differences therefore may have not been great enough to cause any difference in feed preference and intake. It must also be noted that the pattern of feed preference and intake observed during Trial 2 for the DMI and %DMI did not correspond with the differences observed in the colour attributes. It is therefore not clear whether the colour of the feed influenced the feed preference and intake of the birds, but it appears that some other factor apart from colour may have had a more important influence for determining feed preference. This is in contrast to the findings of Bubier *et al*. (1996), who provided strips of insulation tape of different colours (green, white, red, blue, yellow and black) to chicks and found that the green tape produced the greatest pecking response. This can be related to the herbivorous nature of the ostrich in the wild. The second colour of preference was white, which can be related to the coprophagy of adult dung, which is usually accompanied by white urate deposits. However, the results obtained in this study correspond with the results of Brand *et al*. (2008a), who found that while chicks showed a preference for green plastic strips they did not distinguish between feeds of different colours. In addition, Kruger (2007) found that chicks preferred a control diet (untreated pre-starter mash and light brown in colour) to

artificially coloured feeds. The question now arises whether ostriches base their choice of feed selection on the colour of the feed and may therefore warrant further research.

According to Forbes and Covasa (1995), feed intake in a free-choice feeding system does not only depend on the metabolic requirements or physiological state of the bird, but also on factors such as previous experience and social interactions. They advise exposing pullets to all the grains that they may be offered later in life during the rearing period in order to allow them to learn their nutritional characteristics. Rose *et al*. (1986), as well as Forbes and Covasa (1995), also suggested that the type, form and nutrient content of the feed has a profound effect on diet selection. Factors such as trough design, position of the trough, breed, sex, management and genetics can also contribute to determining which diet is selected by the birds. In this study, lupins were the only component of the diet to which the birds had not been previously exposed. In addition, all other possibly influential factors were kept constant and were the same for all the paddocks, reducing the risk of them influencing diet selection.

5.5. CONCLUSION

The results of this study indicate that soybean oilcake meal can be replaced in the diets of grower phase ostriches by sweet lupin inclusion levels of up to 30% without any significant effect on feed selection. The tendency of the birds in Trial 2 to discriminate to some extend against the 15% and 30% bitter lupin diets may warrant further research. Furthermore, it is important to remember that the inclusion of sweet and bitter lupins were only evaluated up to 30% and that replacing soybean oilcake meal beyond this inclusion level could possibly result in a decreased feed intake. Lupins are cost-competitive with multiple other protein sources, making them a widely used raw material in livestock feeds. Results from this study may assist in establishing a potential market for lupins as well as improving the profit margins of ostrich farmers and the local grain legume industry.

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CHAPTER 6

GENERAL CONCLUSIONS AND FUTURE PROSPECTIVES

Ostrich farming has been practiced in South Africa for more than 150 years, and despite continued challenges, it remains a rapidly growing industry. However, production costs are high, with feed being the largest expense in an intensive ostrich production unit (*ca*. 75%). Furthermore, with the growing global population, the decrease in available cropland and the occurrence of more frequent droughts, there is an increasing demand for protein sources. This has resulted in the currently available protein sources experiencing more pressure and becoming more expensive leading to an increasing need for alternative protein sources. Lupins have been identified as a possible alternative, locally produced plant protein source that can be successfully included in the diets of both ruminants and monogastric animals. In some cases it can also replace traditional protein sources such as soybean oilcake meal. However, lupins can only be included up to certain levels to be utilised efficiently and to prevent undesirable effects. Further research to determine the optimal inclusion levels for effective utilisation, the sustainability of this protein source and the available resources for cultivating lupins locally, is required. This study investigated the nutritional potential of lupins to replace soybean oilcake meal in ostrich diets and will contribute to the expansion and improvement of the mathematical optimisation model.

Chapter 3 focused on the effect on the production and slaughter traits of slaughter ostriches when sweet lupins were included in the diet at different inclusion levels. No differences were found between the treatment diets for the live weights, dry matter intake (DMI), average daily gain (ADG) and feed conversion ratio (FCR) at the end of each feeding phase. However, there was a tendency for the birds on the 15% (75LD) sweet lupin inclusion level to have higher ADGs and live weights at the end of the starter phase. No significant differences were found for the dressing percentage and the weight of the big drum muscle of the birds, although birds fed the 50LD and 75LD tended to have the heaviest cold carcasses. The birds receiving the 50LD also had (P = 0.05) heavier thighs than those on the other diets. It can be concluded that soybean oilcake meal can be replaced by up to 15% (75LD) sweet lupins in slaughter ostrich starter diets, and up to 30% (100LD) in grower and finisher diets, without any significant detrimental effects on the production and slaughter traits.

In chapter 4 the quality of the three ostrich end products was evaluated post-slaughter. No differences were found for the marketable feather classes or for most of the leather quality characteristics. However, it was observed that birds on the 25LD had the thickest skins. This corresponds with the end weights of the birds as, although not significant, the birds on the 25LD had the highest end weight. A tendency was found for birds receiving the 25LD and 75LD to have

the highest pinhole densities at locality 4 (the lower flank). However, the number of pinholes is counted by the human eye and is difficult to accurately determine, which could possibly affect the end results. No significant differences were found for the moisture, crude protein and ash content of the meat, but the intra-muscular fat content was influenced by the sweet lupin inclusion in the diet and this should be investigated further. It can be concluded that the sweet lupin inclusion levels evaluated in this study had little influence on the feather classes, leather traits and the meat composition of the birds measured.

The feather yields of the birds in this study were slightly below values found in previous studies. Furthermore, the feathers were not harvested at six to eight months of age, as they were still green feathers at this time. Research on feather growth and quality has been limited due to this product contributing only a small portion (~10%) of the total income of a slaughter bird. However, it may be beneficial to the industry to determine the importance of feather clipping and at what stage they should be clipped when slaughtering at 12 to 14 months of age if the feathers are not yet ripe at six to eight months.

Furthermore, the high mortality rate of ostrich chicks is a constant struggle in commercial ostrich production units and was also a problem in this study. Therefore, further research on improving ostrich chick survival is needed. Additional research in the field of ratite behaviour may help producers better understand these birds and adapt their handling procedures so that minimal stress is placed on the bird. This may also help lower bird aggression and ultimately reduce skin damage and bruising, which will result in higher product returns.

The results obtained in this study were generally similar to the production standards derived from previous growth studies; however, it is important to note that sweet lupins were only evaluated up to an inclusion level of 20% in the starter phase and 30% in the grower and finisher phases. Including sweet lupins in ostrich rations beyond the levels evaluated may result in findings differing from the results reported in this study. However, ostrich producers can be assured that including sweet lupins in the diets of slaughter ostriches at the inclusion levels tested will yield similar quality returns as for the standard commercial slaughter ostrich diets, which are formulated with soybean oilcake meal as a protein source.

Chapter 5 assessed the effects of sweet (Eureka) and bitter (SSL10) lupins on the feed preference of growing ostriches by monitoring the DMI per bird per day for each diet. It was concluded that sweet lupins can be included in the diet up to 30% without any significant effect on feed selection. The tendency (P = 0.11) of the birds to discriminate to some extend against the 15% and 30% bitter lupin diets may warrant further research. Once again it is important to remember that the sweet and bitter lupins were only evaluated up to 30% and that replacing soybean oilcake meal above this inclusion level could possibly result in a decreased feed intake. Although this is not

scientifically proven, higher lupin inclusion levels may result in higher alkaloid levels which will result in the feed being unpalatable which may reduce feed intake.

Ostriches perform well on high-fibre diets relative to other monogastric animals (poultry and pigs), thus making lupins a valuable alternative to soybean oilcake meal in ostrich rations. Their hindgut fermentation ability most likely enables them to utilise lupins, which have a relatively high fibre content, more efficiently. Furthermore, lupins are cost-competitive with multiple other protein sources, making them a widely used protein and energy source in livestock feeds. Currently the price of sweet and bitter lupins is 56% and 28%, respectively, of the price of soybean oilcake meal. This makes the use of lupins a viable economic alternative, as it lowers input costs without compromising any of the production and slaughter traits or the quality of the ostrich end products.

The results from this study will contribute to the continued development of the mathematical optimisation model of the nutrient requirements of ostriches as developed by Gous and Brand (2008). The ostrich industry will benefit from this model as producers will eventually be able to predict what will happen to each of the ostrich end products when the diet is altered and be able to formulate least-cost diets that are closer to the needs of the birds. The results will also contribute to the currently limited knowledge of the nutritional value of sweet and bitter lupins as raw materials in ostrich rations and consequently improve the economical production of ostriches.

These findings may also assist in creating a potential market for locally produced protein sources, such as sweet and bitter lupins, in South Africa and broaden our knowledge of the potential of this raw material as a feed ingredient for animals. Other plant protein sources such as canola oilcake meal and full-fat canola may also warrant further research as potential alternatives to soybean oilcake meal, which is currently commonly used within the industry. The incorporation of locally produced plant protein sources in the diets of slaughter ostriches will not only reduce the high input costs for nutrition but will also be beneficial for the local grain legume and oilseed industry.

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APPENDIX 1

The composition of the vitamin and mineral premix used in the four ostrich feeding phases (pre-starter, starter, grower and finisher) formulated per ton of feed.

Ingredients		Stage of growth	
(Composition per unit of premix)	Units	Pre-Starter & Starter	Grower & Finisher
Vitamin A	IU	15 000 000	12 000 000
Vitamin D3	IU	4 000 000	3 000 000
Vitamin E	mg	60 000	40 000
Vitamin K3 stab	mg	3 000	3 000
Vitamin B1	mg	5 000	3 000
Vitamin B2	mg	10 000	8 000
Vitamin B6	mg	8 000	6 000
Vitamin B12	mg	100	100
Niacin	mg	100 000	80 000
Pantothenic Acid	mg	15 000	12 000
Folic Acid	mg	3 000	2 000
Biotin	mg	300	200
Choline	mg	800 000	600 000
Magnesium	mg	50 000	50 000
Manganese	mg	120 000	120 000
Iron	mg	30 000	25 000
Zinc	mg	120 000	80 000
Copper	mg	8 000	8 000
Cobalt	mg	300	100
Iodine	mg	2 000	1 000
Selenium	mg	300	300

**RECOMMENDATION*: To make half ton of feed divide premix pack into two parts.