NUTRITIVE VALUE AND EVALUATION OF APPROPRIATE FEEDING STRATEGIES OF GRAIN SORGHUM VARIETIES FOR POULTRY IN TANZANIA

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EXTENDED ABSTRACT

In Tanzania, maize scarcity, high cost and competitiveness between humans and other livestock has necessitated the search for alternative energy sources for poultry. In view of this, four studies on the assessment of the suitability of commercially available Tanzanian grain sorghum varieties (GSV) for poultry and evaluation of appropriate feeding strategies of diets containing GSV were conducted. In the first study, a cross section survey was conducted in selected food markets of Singida, Dodoma and Morogoro regions to identify types of available commercial GSV whereas in the second study, assessment of nutritive values of identified GSV was carried out. In the third and fourth study, two feeding experiments to evaluate the effectiveness of using final sugar molasses (SCM) and *Moringa oleifera* leaf meal (MOLM) as additive in GSV based diets on performance of growing chicks and layer chickens and egg quality characteristics and consumers' preferences were carried out. Semi-structured questionnaire and personal observation guided by checklist were used to collect information on type, identity and source of GSV. The identified GSV were analyzed for proximate and anti-nutritional components, minerals, some amino acids, metabolizable energy (ME_n) and starch using standard procedures in the second study. GSV collected from local food markets, were ground and packed in nylon bags for use in the feeding experiments. Fresh Moringa leaves were harvested dried under the shed, ground through hammer mill to make MOLM. Molasses was obtained from Mtibwa Sugar Company. Other feed ingredients were obtained from local livestock feed dealers. 2x3 factorial experiment in which two GSV for high tannin sorghum (HTS) and low tannin sorghum (LTS); three supplementary strategies for non-supplemented GSV based diets (CTL) and 10% DM- SCM and

10% DM-MOLM (MOL) supplemented GSV based diets to make six dietary treatments for the feeding experiments. The data of the second study was analyzed using descriptive statistics whereas for the third and fourth study the General Linear Model Procedure was used. The results showed that out of the twelve Tanzanian commercial GSV identified, 75% were predominately landraces with local identity.58% of GSV had white coat colour and were sourced from Singida and Dodoma region. The overall chemical composition for GSV was found to be dry matter (DM) (88.3% ± 0.44), metabolizable energy (ME_n) (13.8MJ/KgDM±0.39), ether extract (EE) (3.2%DM±0.46) and crude protein (CP) (11.1%±0.63). In addition, GSV contained a range of high crude fiber (CF) (1.8-6.8%DM), ash (1.8-6.9%DM) and condensed tannins (2.1-5.8%DM) as leucocynidin equivalent and low macro/micro minerals and essential amino acids., The results did not show any significant correlation between coat colour and amount of condensed tannins in GSV. The results of the present study showed that use of HTS as a main source of energy led to poor dry matter intake (DMI), lymphocyte, and oesinophils and monocyte ratios for growing chicks whereas poor DMI, feed conversion and egg production were observed in layer chickens but produced more desirable eggs in terms of aroma, flavour and yolk colour after being stored for long time. The LTS diets had better DMI, lymphocyte, oesinophils and monocyte ratios and feed conversion for growing chicks whereas better feed conversion and egg production were observed in layer chickens but relatively produced undesirable eggs in terms of aroma, flavour and yolk colour after being stored for long time. The results further showed that supplementation of GSV based diets with final SCM led to increased leg abnormality incidences (LAI), paleness of the bird's body parts and decreased lymphocyte and oesinophils ratios for growing chicks. In layer chickens decrease in egg production and impairment of feed conversion, egg aroma, flavor and yolk colour when final sugar cane molasses was added to GSV based diets were noted. Addition of MOLM to GSV based diets, prevented occurrence of LAI and improved colour of the bird's body parts; monocyte, lymphocyte and oesinophils ratios and feed conversion for growing chicks. Egg production, feed conversion, Roche egg yolk color scores (RYS) and consumers' yolk colour score index (YSI) were improved by the addition MOLM in GSV based diets for layer chickens. It can be concluded that Tanzanian commercial GSV have high feed component and energy content showing their potential in poultry feeding but their utilization could be limited by presence of high anti-nutritional factors and low content minerals and amino acids. Moreover, HTS has relatively lower feeding values than LTS in poultry feeding may be due to the negative effects of tannins which impair the utilization of nutrients though they may produce relatively more desirable egg in terms aroma and flavour qualities when stored for long time. However, LTS has higher feeding value for poultry due to nutrition adequacy although may cause to leg abnormalities in chicks if it contains high phytates and may lead to unpleasant egg aroma, flavour and yolk colour when stored for long time. It can further be concluded that utilization of MOLM additive to GSV based diet for poultry improve feeding value of HTS and LTS in layer chickens due to its ability to compensate deficient minerals, pigments, energy, amino acids and proteins. But, use of SCM additive to GSV based diet for poultry worsens HTS and LTS feeding values due to the negative its effects in utilization of nutrients associated with its ability to increase rate of passage in gastro-intestinal tract. Following findings of this study, partial utilization of Tanzania GSV as a source of energy in poultry feeding is recommended but it's fully use should be accompanied by supplementation of synthetic minerals, pigments and amino acids or strategic feeding options. Moderate use of HTS in layer chickens' diets is suggested but fully utilization should require supplementation of synthetic pigments, minerals and amino acids and energy sources or strategic feeding options for health chicks and production of desirable eggs. Fully utilization of LTS in layer chickens' diet is highly recommended but requires supplementation of synthetic pigments and minerals for production health chicks and desirable eggs. Addition of up to 10%DM- MOLM level for improving HTS and LTS feeding value for layer chickens is recommended whereas the use of 10%DM- final SCM level is not suggested. Further research is recommended to evaluate economics of using MOLM additive to GSV based diet for layer chickens and nutritive value of different GSV in Tanzania rural.

DECLARATION

I, JOHN THEODOSIAS KAIJAGE, do hereby declare to the Senate of Sokoine University of Agriculture that this thesis is my own original work done within the period of registration and has neither been submitted nor being concurrently submitted in any other institution.

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DEDICATION

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LIST OF PAPERS AND MANUSCRIPTS

This thesis is based on the following studies (papers and manuscripts), which will be referred to in the text by their roman numerals:

Paper I: Kaijage, J. T., Mutayoba, S. K. and Katule, A.

Chemical composition and nutritive value of Tanzanian grain sorghum varieties.

Status: Published in *Livestock Research for Rural Development 26(10).* **Retrieved:** November 2, 2014 from http://www.lrrd.org/lrrd26/10/kaij26177.htm

Paper II: Kaijage, J. T., Mutayoba, S. K., Katule, A. and Kakengi A. M. V. Relative effects of *Moringa oleifera* leaf meal and molasses as additives in grain sorghum based diets on performance of growing chicks in Tanzania.

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Manuscript: Paper III: Kaijage, J. T., Mutayoba, S. K. and Katule, A.

Effect of *Moringa oleifera* leaf meal and molasses additives in grain sorghum based diets on haematology and serum bio-metabolites for growing chicks.

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Paper IV: Kaijage, J. T. Mutayoba, S. K. and Katule, A.

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Moringa oleifera leaf meal and molasses as additives in grain sorghum based diets; effects on egg quality and consumer preferences.

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ABBREVIATIONS AND SYMBOLS

%	Percentage
^0C	Degree Centigrade
ADF	acid detergent fiber
ASI	aroma score index
CF	crude fiber
CP	crude protein
DASP	Department of Animal Science and Production
DM	dry matter
DMI	dry matter intake
ECC	Egg cholesterol concentration
EE	ether extract
EMP	egg mass production
ESI	egg shell thickness
et al	and others
FAO	Food and Agricultural Organization
FCR	feed conversion ratio
GAS	general acceptability egg scoring index
GLM	General Linear Model
HTS	high tannin sorghum
LAI	leg abnormality incidences
LTS	low tannin sorghum
ME_n	metabolizable energy

MLFD	Ministry of Livestock and Fisheries Development
MOLM	*Moringa oleifera* leaf meal
PAS	Percent albumen solids
PEA	Percent egg albumen
PES	percent egg shell
PEY	Percent egg yolk
PYS	Percent egg yolk solids
RBC	red blood cells
RYS	Roche yolk colour score
SAS	Statistical Analysis Systems
SCM	Sugar cane molasses
SEM	Standard error of mean
SUA	Sokoine University of Agriculture
TASI	taste score index
TSI	total egg scoring index
Vs	Against
YSI	egg yolk score index

CHAPTER ONE

1.0 GENERAL INTRODUCTION

1.1 Background Information

In Tanzania, poultry production is one of the major agricultural activities in recent years but has been growing moderately in both traditional and commercial sub-sectors (FAO, 2005). The moderate increase in the annual per capita production of poultry products (eggs and meat) does not meet the current domestic demand of poultry products (FAO, 2005) and has resulted into a deficit in trade balance (FAO, 2005a). Moreover, in recent years, in Tanzania an increasing demand of poultry products (poultry meat and eggs) has prompted the Government to promote commercial poultry production so as to minimize import of poultry products (FAO, 2005a). The growing demand for poultry products is a result of increased tourist hotels, middle class population, supermarkets and mining industry. One of the challenges of poultry production and productivity is the availability of quality poultry feeds at affordable cost. Maize is the major energy source in poultry diets in Tanzania but at the same time it is a staple food for most people. These scenarios have led to increased competition for maize between humans and livestock feed manufacturing industries (Yegani, 2009). This competition in most cases results in increased maize price and increased cost of poultry feeds (Tangedjaja, 2011; Mutayoba *et al.* 2011) and is more distinct in areas where availability of maize is a problem due to unreliable rainfall and other agronomical conditions such as soil fertility (Olugbemi *et al.* 2010). The increasing scarcity and high cost of maize used as the source of energy consequently slow down poultry production and the expansion of the poultry industry. This has compelled researchers and producers to look for alternative feed ingredients that are relatively cheap, less competitive and locally available (Mutayoba *et al.* 2011). Amongst such source that have shown potential to replace maize as source of energy in poultry feeds are the grain sorghum varieties (GSV).

1.2 Potential of Utilizing GSV for Poultry in Tanzania

The average annual production of grain sorghum in Tanzania is estimated to be over 700,000mt and it's the highest in the Southern African countries. Sorghum is the second most widely grown cereal crop in the country and has a wide genetic diversity. It is mostly grown by small peasant farmers in small plots under subsistence production systems in the semi-arid regions of Dodoma, Singida, Shinyanga, Mwanza, Mara, Lindi and the drier parts of Morogoro (Minde and Mbiha, 1993 and Roharbach and Kiriwaggulu, 2007). More than 95% of grain sorghum produced is utilized at home and only about 2% reach the formal markets due to lack of organised commercial markets (Roharbach and Kiriwaggulu, 2007). Thus, promotion of utilization of grain sorghum for poultry could create markets which could lead to increased production. Sorghum plant can adapt and withstand the effects of climate change better than maize (Roharbach and Kiriwaggulu, 2007) as it can grow in marginal soils and yield more than maize in drought years. It is for these reasons that many farmers are replacing maize with sorghum in areas where rainfall has been declining due to climate change. Grain sorghum grows quickly and requires less labour and is more resistant to pests and diseases compared to maize.

The nutritive value of grain sorghum is comparable to maize in terms of chemical composition (FAO, 1995; BSTID-NRC, 1996). Starch is the main component of grain sorghum, followed by proteins, non-starch polysaccharides (NSP) and fat. The macromolecular composition of sorghum is similar to that of maize and wheat (BSTID-NRC, 1996). The NSP in grain sorghum is mainly constituted by arabinoxylans and other glucans (Hatfield *et al.* 1999). Grain sorghum also contains non-carbohydrate cell wall polymers as lignin with proportions constituting up to 20% of total cell wall materials (Hatfield *et al.* 1999). The protein content of whole grain sorghum ranges between 7 and 15% (FAO, 1995; Beta *et al.* 1995). Sorghum proteins are divided into albumins, globulins, kafirins (aqueous alcohol-soluble prolamine), and cross linked kafirins and glutamines (Jambunatan *et al.*1975). The kafirins constitute about 50-70% of the proteins (Doudu *et al.* 2003). However, there is a wide variability between varieties in respect to the levels of protein in sorghum (Reddy and Eswara, 2002). The fat content in grain sorghum (mainly present in germ) is rich in fatty acids such as linoleic- (49%), oleic-(31%), palmitic-(14%), linolenic (2.7%), stearic (2.1%), similar to maize (Glew *et al.* 1997). Sorghum is a good source of vitamins notably the B and fat soluble vitamins and is also a good source of more than 20 minerals particularly phosphorous, potassium, iron and zinc (Anglani, 1998). The adaptive features of sorghum crop to climate change and its wide distribution, substantial production and nutritive value of grain sorghum demonstrate its potential in replacing maize in poultry feeding in Tanzania.

1.3 Factors Affecting Utilization of GSV for Poultry

Some GSV contain anti-nutritional factors that lower the feeding value for poultry. These include tannins and phytates and inherent physiochemical characteristics (Roharbach and Kiriwaggulu, 2007; Butler *et al.* 1992). Tannins and phytates are major nutritional concerns for GSV in poultry diets. Tannins impart astringent taste which affects palatability and consequently feed intake and body growth (Butler and Rogler, 1992). Tannins also bind endogenous and exogenous protein, cellulose, hemicelluloses, pectin, phytates and minerals to form indigestible complexes (Van Soest, 1994). Therefore, high tannin levels in sorghum adversely affect digestibility of protein and carbohydrates, reduces growth, feed efficiency, Metabolizable energy and bioavailability of amino acids. At levels of more than 2% of tannic acid in the diet was found to decrease egg production, egg weight, shell quality, yolk colouration and increase egg mottling (Sell *et al.* 1983; Blakelee and Wilson, 1979; Jacob *et al.* 1996; Issa *et al.* 2007). Grain sorghum also contains phytates, a mixed salt of phytic acid. Phytates have the ability to bind phosphorous (P) and form complexes with protein and minerals which impair their utilization (Bryden *et al.* 2007). The total P content in grain sorghum ranges between 3-4g/kg whereas P bound by phytates ranges from 2.1 to 2.4g/kg and this in most cases not available to poultry (Selle *et al.* 2003). Also, physiochemical characteristics of GSV may negatively contribute to their feeding values. Sorghum starch granules are surrounded by a protein matrix which can limit access of enzymes (Benmoussa *et al.* 2006). Other factors important to the energy value of sorghum include channels or pores on starch granules that are sites for enzyme entry (Benmoussa *et al.* 2006), granule size, starch-lipid complexes and kafirins content (Cao *et al.* 1998). In addition, waxy starch is more digestible than non-waxy (conventional) sorghum. Hard grain sorghum has cross-linked kafirins which negatively affect digestibility (Abdelrahaman and Hoseney, 1984). The GSV with vitreous endosperm are less digestible because they contain more proteins, kafirins and disulfide bonds than floury endosperm which has more soluble proteins (Bryden *et al.* 2009). Due to the foregoing, different strategies to improve the feeding value of GSV are required in order to increase and maximize its potential in poultry feeding. Most of the earlier studies in improving grain sorghum were based on reducing tannin content. However, the adoption of these mechanisms has not been forthcoming due to being either too expensive or not practical under practical production conditions. It has also been noted that some of the mechanisms only result in partial improvement of the feeding value. It was therefore thought that supplementation with cheap and locally available materials might improve the feeding value of grain sorghum based diets for poultry. *Moringa oleifera* leaf meal (MOLM) and final sugar cane molasses (SCM) seem to be relatively cheap, readily and technically applicable under local condition. Both MOLM and SCM have been used in improving productivity of poultry when used with other feedstuffs thus they could be utilized as strategic options for improving GSV based diets for poultry.

1.4 Potential of MOLM and Final SCM in Improving Feeding Value of GSV

Moringa oleifera is a perennial plant which grows in most tropical countries, including Tanzania (Baumer, 1983; Makker and Becker, 1996; Optima Africa, 2000), and is drought tolerant and can survive under harsh conditions (Duke, 1987) and its annual dry matter yield is high compared to other forages in the tropics (Becker and Marker, 2000). The leaves of *M. oleifera* are a good source of essential nutrients such as Beta-carotene, vitamin C (Ascorbic acid), iron, free leucine, vitamins and minerals such as calcium content and phosphorous, sulphur containing amino acids and metabolizable energy (ME_n) compared with other tropical plants (Chawla *et al.* 1988; Makkar and Berker, 1996; Makkar and Becker, 1997; Price, 2000). *M. oleifera* leaves contain insignificant levels of anti-nutritional factors and toxic materials and are high in naturally occurring plant phyto-chemicals compared to other shrubs and browse legume leaves (Makker and Becker, 1997; Fahey, 2005). Previous studies have shown that incorporating MOLM in layer diets had no detrimental effects on egg production and feed conversion ratio (Kakengi *et al.* 2007; Abou-Elezz *et al.* 2011). Furthermore, inclusion of MOLM in broiler and layer diets led to improved weight gain, carcass quality and egg quality characteristics and reduced blood triglycerides, enhanced immune response, promoted metabolism and increased natural defenses of the body (Caceres *et al.* 1991; Kaijage, 2003; Yang *et al.* 2006 and Olugbemi, 2009). Therefore, the present review show that MOLM has high potential to alleviate the negative effects of anti-nutritional factors such tannins and phytates in GSV based diets due to its ability to compensate bound minerals, proteins, essential amino acids and energy.

Final SCM is readily available and produced in large amounts from the sugar manufacturing industries in Tanzania. The final SCM is found in liquid or dried form and exerts a tonic effect, induces palatability and can eliminate dustiness in poultry feeds. Final SCM contains relatively large amount of total sugars or carbohydrates, NFE and minerals. The utilization of molasses in poultry feeding is well documented (Keshavarz *et al.* 1980). The inclusion of SCM in maize based diets for layer chickens led to improved laying performance and egg mass production

(Sharma and Paliwal, 1973). The inclusion of SCM in wheat based diets for laying pullets improved growth and laying performance (Rahman *et al.* 1991) The supplementation of SCM in sand or wheat straw based diets for broiler chick diets gave better performance compared to those chicks fed wheat straw only (El-Sagheer, 2006). Also, broilers fed 15% SCM had better performance in terms of growth, final weight, nutrient digestibility compared to animals that were fed lower amounts or diets without SCM (Njidda *et al.* 2006). Broilers fed 11 and 12% molasses in the starter and finishing rations respectively in substitution for sorghum grain had similar performance to birds fed grain sorghum based diet (control). However, the inclusion of 39 and 42% SCM in the starter and finishing rations significantly decreased feed intake, increased live weight, feed efficiency and dressed carcass percentage (Rahim *et al.* 1999). The supplementation of 5-7ml per litre of molasses in drinking water for broiler chicks increased live weight gain, enhanced haemopoiesis, ameliorated the effect of oxidative stress induced by heat stress (Habibu *et al.* 2014). However, the effectiveness of MOLM and SCM in improving feeding value of grain sorghum for poultry is not clearly understood.

1.5 Problem Statement and Justification

1.5.1 Problem statement

In Tanzania, high price, scarcity and competition for maize between human and livestock feed processing industries has resulted in the production of low quality and expensive poultry feeds. This scenario has necessitated the need to look the possibility of using GSV as an alternative energy source in poultry diets. However, sorghum is not preferred by commercial livestock feed processors and farmers because it is viewed as an inferior feed ingredient compared to maize and that it could negatively affect poultry performance (Rohrabacher and Kiriwaggulu, 2007). The feeding value of all GSV is perceived to be sub-optimal for poultry and low in energy, protein values and amino acids. In addition, all Tanzanian GSV are known to contain high anti-nutritional factors such as tannins (Rohrabacher and Kiriwaggulu, 2007) which could in way cause mortalities and impair performance of birds. Thus, the utilization of grain sorghum for poultry has been below 10% of the total grain sorghum production (MAC, 1998; Rohrabacher and Kiriwaggulu, 2001). The differences in feeding values of GSV are not well understood by farmers and feed processors, hence identifying these differences and improving feeding value through strategic options could eventually increase its utilization in poultry diets.

1.5.2 Justification

Promotion of the utilization of grain sorghum to mitigate the scarcity of maize as a main source of energy and a need to improve Tanzanian poultry feed processing industry triggered the need to understand the nutritive values of commercially available Tanzanian GSV. This information is vital to enable selection of suitable GSV for poultry feeding. The suitable GSV could be promoted and unsuitable ones improved for poultry. However, in Tanzania there is scanty information about the nutritive value of different commercial GSV. Few studies done have concentrated mainly on GSV that are cultivated in rural areas for food security and their nutritive value for humans (Minde and Mbiha, 1993; Mafuru *et al.* 2007). On the other hand, utilization of GSV for poultry require a feeding strategic options that can improve feeding value of unsuitable GSV for poultry. However, different suggested strategic options for improving GSV have not been forthcoming in the Tanzanian context (Sell and Rogler, 1984; Armstrong *et al.* 1974; Chavan *et al.* 1979; Douglas *et al.*

1988; Douglass *et al.* 1990; Wyatt, *et al.* 1997; Cramer *et al.* 2003; Mikkelson *et al.* 2008) because are either expensive or not easily handled and therefore not practical. This phenomenon triggered the need to look for cheap, practical, locally available and viable options. Use of MOLM and SCM could be potential strategic options for improving feeding value of GSV for poultry. However, there is scanty documented information on the effectiveness of MOLM and SCM in improving feeding value of GSV for poultry. Few studies carried out focused more on inclusion of MOLM and SCM in maize based diets for poultry. Based on these gaps a study was formulated to evaluate the nutritive value of Tanzanian GSV for poultry and compare effects of SCM and MOLM as additive to GSV based diets for growing and layer chickens as well as its effects on egg quality characteristics and consumer preferences. The findings of this study form a basis to determine the suitability of different Tanzania commercial GSV for poultry. The study further forms the basis for recommending the most suitable strategic option between SCM and MOLM. The results of this study will also be important for policy makers, grain sorghum producers; poultry feed processors and poultry farmers for promoting cultivation and utilization of GSV for poultry. This could increase poultry productivity and eventually contribute to alleviation of poverty.

1.6 Objectives

1.6.1 Overall objective

The overall objective of the study was to improve feeding value of Tanzanian GSV for poultry by evaluating its nutritive value and the effectiveness of MOLM and SCM in improving their feeding values.

1.6.2 Specific objectives

The specific objectives were to:

1) identity available Tanzanian commercial GSV and determine their bio-chemical composition.
2) determine and compare the effects of SCM and MOLM additives to GSV based diets on performance of growing chicks.
3) determine and compare the effects of SCM and MOLM additives to GSV based diets on performance of layer chickens.
4) identify and contrast the effects of SCM and MOLM additives to GSV based diets on egg quality characteristics and consumers' egg quality preferences.

1.7 Research Hypothesis

i) Different Tanzanian commercial grain sorghum varieties found in food markets differ in origin and biochemical composition.

ii) Uses of HTS and LTS as main source of energy in poultry diets differ in feeding values and inflict diverse effects on performance of growing chicks and layer chickens and egg quality characteristics.

iii) Uses of final SCM and MOLM have comparable feeding values and impose similar effects on performance of growing chicks and layer chickens and egg quality characteristics.

1.8 Organisation of the Thesis

This thesis is arranged in eight chapters. The first chapter contain general introduction. The introductory part highlights background information for poultry sub- sector in Tanzania. The production trend, potentials and challenges of poultry production and productivity are highlighted. Feeding as a major challenge for poultry productivity is pointed out. Limitation of maize as a main source of energy for poultry is highlighted. Potentials and limitations of utilizing GSV for poultry are given. Strategies for improving GSV for poultry and their limitations in Tanzania context are shown. Potentials of MOLM and SCM as cheap strategic option for improving GSV feeding are provided. In this chapter also the problem statement and justification of the study are described. This chapter also states the objective and hypothesis behind the study. The chapter two contains the general methodology of the study. In this chapter materials and methods used in the study are stated. The chapter also shows the organization of the thesis and ends with a list of references to support information highlighted in chapter one and two.

Chapter three to seven in the thesis contain four published papers and one manuscript sent to peer reviewed scientific Journal. Each publication is a chapter in this thesis. The published papers are (Paper I, II, IV and V) and a manuscript publication is (Paper 111). Chapter eight contains general discussion in which the key findings of the study are synthesized. This chapter also gives general conclusive remarks of entire study. Finally, the chapter gives recommendations of the entire study.

CHAPTER TWO

2.0 MATERIALS AND METHODS

To achieve specific objective 1, a cross section survey was carried out in nine food markets located in the urban and peri-urban areas of Singida, Dodoma and Morogoro regions of Tanzania. In each market all grain sorghum retailers and whole sellers and some key informants were interviewed. A semi-structured questionnaire and personal observation guided by a checklist were used to collect information on type, identity and source of GSV. Samples of identified GSV were collected and analyzed for proximate and anti-nutritional components, minerals, some amino acids, ME_n and starch. Dry Matter (DM), Crude Protein (CP), Ether Extract (EE) and ash in the sorghum grain samples were determined using the (AOAC, 1990) standard methods. The minerals in grain sorghum samples were determined using atomic absorption spectro-photometric method described by (AOAC, 1990). ME_n were determined by a formula as expressed by (NRC, 1994). Condensed tannins in grain sorghum were determined using Butanol/ HCL methods as described by Nitao *et al.* (2001). Total phenolic compounds were determined using the procedure described by (Ciorol and Dumitriu, 2009). Phytates were determined based on an iron to phosphorous ratio of 4:6 using the method as described by (Mustafa *et al.* 2003). The results from the study grave comparative and general information on suitability of Tanzanian commercial GSV for poultry.

To achieve specific objective 2, a feeding experiment was conducted at the poultry unit of the Department of Animal Science and Production (DASP), Sokoine

University of Agriculture (SUA). Two GSV based on being HTS and LTS, locally known as Serena and Mbangala respectively were purchased from local food market, ground to pass through 2mm with a hammer mill to produce grain sorghum meal and stored in clean nylon bags. *Moringa oleifera* leaves were harvested from trees in DASP compound. Moringa tree branches were cut, spread out and dried under the shade for a period of 3 to 4 days, threshed carefully before milling and ground with hammer mill and stored in nylon bags. Final SCM was purchased from Mtibwa sugar processing company, packed and stored in plastic containers. Other feed ingredients used such as fish meal, limestone, salt, bone meal and sunflower seed meal (SSM) in this study were purchased from local livestock feed dealers. The experimental diets were formulated to meet or exceed nutrient recommendations for growing chicks. The proximate composition, ash, phosphorus and calcium for feed ingredients and diets were determined according to the methods of AOAC, (1990). Metabolizable Energy (ME/kcal/kg) content of feed ingredients and the experimental diets were estimated by prediction equations as expressed in NRC, (1994) and MOLM, ME/Kgcal was estimated by prediction equation established by Carpenter and Clegg, (1956). Condensed tannins in GSV and MOLM were determined using butanol/HCL method as described by Nitao *et al.* (2001).

Two hundred and seventy (270) day old bovan hybrid egg strain chicks, obtained from Songwe hatchery in Mbeya City Council, Tanzania were randomly allotted to three replicates in six dietary treatments arranged in a 2x3 factorial design for two GSV (HTS and LTS based diet) and three supplementary strategies (SPS) (non-supplemented GSV (0%) (Control; 10% dry matter (DM)-SCM and 10%DM-MOLM supplemented GSV based diet) to make six dietary treatments. Birds were randomly allocated to treatment groups of 45 birds each. Each treatment group was further subdivided in 3 replicates of 15 birds each. Each treatment replicate was randomly placed in a single pen of a poultry house for eight weeks. Birds were wing banded and weighed individually immediately obtain individual initial bird weight and thereafter on a weekly basis for body weight changes. Abnormalities and mortalities were noted and recorded daily. Feed offered and leftovers were recorded daily to determine daily feed intake (DFI). Daily weight gain (DWG) was calculated as individual average weight gain over a period of time. The DFI (g/bird) over DWG (g/bird) was used to calculate FCR. Birds with abnormal legs were recorded daily. Leg abnormality incidences (LAI) were calculated as the ratio of birds with leg abnormalities to total number of birds available x 100.

After eight (8) weeks on the feeding experiment, two birds were randomly selected from each treatment replicate; six birds per treatment and a total of 36 birds for the six dietary treatments were randomly placed in battery cages for evaluation of haematology and serum bio-metabolites. A total of 6ml of blood was sampled from each bird by which 2mls were placed into bijon bottle treated with ethylene diamine tetra acetic acid (EDTA) for haematological assay. The remainder (4ml) of the blood sample was allowed to coagulate to produce sera for blood chemistry measurements according to the methods of Okeudo *et al.* (2003). Blood samples were analyzed for total erythrocyte, haematocrit (PCV), haemaglobin (Hb) and differential leucocytes count according to the methods described by Dein, (1984). The coagulated blood was subjected to standard method of serum separation of the harvested total serum protein (TSP) using Biuret method end point whereas albumin concentration was determined by the Bromocresol Green (BCG) Dye method described by (Peters *et al.*

1982). Globulin (Gb) concentration was computed as the difference between total protein and albumin concentrations. Glucose concentration in serum was determined by the Glucose oxidase/peroxidase method whereas cholesterol concentration in serum samples was determined by chod-pap method. The results from this experiment showed comparative information on the effect using HTS and LTS without supplementation and the effect of supplementing SCM and MOLM on haematology and serum bio-metabolites of chicks.

To achieve specific objective 3, a feeding experiment was conducted at the poultry unit in the Department of Animal Science and Production (DASP) of the Sokoine University of Agriculture (SUA). Two hundred and seventy (270) bovan hybrid layer chickens at 20^{th} week of age were randomly allotted to three replicates in six dietary treatments arranged in a 2x3 factorial design for two GSV for high tannin sorghum (HTS) and low tannin sorghum (LTS) based diet and three supplementary strategies (SPS) (non-supplemented GSV (0%) (Control); 10%DM-SCM and 10%DM-MOLM) supplemented GSV based diet. Birds were randomly allocated to treatment groups of 45 birds each. Each treatment group was further subdivided in 3 replicates of 15 birds each. Each treatment replicate was randomly placed in a single pen in an open ended poultry house for ten weeks. Birds were wing banded and weighed individually immediately to obtain individual initial bird weight. Thereafter, birds were weighed at the end of experimental period. Birds were group fed in each replicate. Feed offered and leftovers were recorded daily for calculating feed intake. Eggs were collected and weighted daily in each replicate group. Feed intake was calculated by taking the difference between the feed offered and residual in each replicate. Feed intake per bird was calculated by subtracting the leftovers from feed and was divided by the number of birds. Eggs were collected and recorded on a daily basis. Eggs were weighed daily using sensitive digital weighing scale. Daily laying percentage was calculated as the number of eggs produced over a period of seven days by dividing the number birds x 100. Average egg weight was calculated as total egg weight divided by the number of recorded eggs. Daily egg mass was calculated as average egg weight multiplying by daily laying percentage. The daily feed intake (g) over daily egg mass (g) was used to calculate the feed conversion ratio (FCR).

Four eggs from each replicate group, (12 eggs per treatment) were randomly sampled after every three weeks for assessment of physical and internal characteristic egg parameters. The egg shape index was calculated and expressed as a ratio of egg length to width. Egg length was measured in centimeter (cm) along the longitudinal axis and the width measured along the equatorial axis using a vernier calliper. Eggshell thickness was measured using a micro-meter gauge. Egg component proportions were weighed and proportions of their weights calculated. The albumen weights were determined by the difference between the whole egg weight and that of the yolk added to the shell weight. The egg component proportions were determined based on egg weight. Egg yolk colour scores were determined using a 15 grade Roche fans (Hoffman Roche Switzerland). The egg, albumen and yolk solids were determined according to the AOAC, (1990) procedure. Egg yolk cholesterol concentration was determined by chod-pap method.

At the end of the feeding experiment, sixty (60) untrained panelists, thirty (30) women and thirty (30) men, were randomly selected for evaluation of consumers' preferences of eggs from different dietary treatments. Five score Hedonic sensory test of 1,2,3,4 and 5 with scoring indices of 1-20, 21-40, 41-60, 61-80 and 81-100% for dislike very much, dislike moderately, neither like or dislike, like moderately and like very much were used. The mean score of studied parameters in each replicated group was calculated by summation of score dividing to the number of panelists. Then the mean score index in each replicate group was calculated by mean score divided by highest score rank x 100. The results from feeding experiment gave information on the effects of utilizing HTS and LTS as main sources of energy on performance of layers and egg quality. Moreover, the experiment showed the influence of using SCM and MOLM as additive on HTS and LTS based diets on performance of layers and egg quality.

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CHAPTER THREE

3.0 PAPER I

Chemical composition and nutritive value of Tanzanian grain sorghum varieties

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Chemical composition and nutritive value of Tanzanian grain sorghum varieties

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Abstract

Two studies were conducted to assess' type, suitability, nutrient composition and anti-nutritional components of commercially available Tanzanian grain sorghum varieties' (GSV) for poultry feeding. In the first study, a cross section survey was carried out in nine food markets located in the urban and peri-urban areas in three regions of eastern central Tanzania to identify available commercial GSV. In the second study proximate, nutritional and anti- nutritional components of GSV were analyzed. Data were analyzed using descriptive statistics. Twelve GSV were identified, most (75%) of them were landraces and few (25%) were improved. Fifty eight percent (58%) of GSV had white coat colour and the rest (42%) had red or yellow. The overall mean for the proximate components, Metabolizable energy (ME_n), starch, amino acids and anti-nutritional factors were expressed as percentage dry matter (%DM). The mean of values for DM (88.3±4.5), crude protein (CP) (11.2% ± 0.68), nitrogen free extract (NFE) 78.0%±2.62, starch 70.7% and (ME_n) (13.2MJ/DM ± 0.39) were high whereas ether extract (EE) (3.15%±0.68) were moderate and phytates (2.42µg/100gDM) were low suggesting high nutritive value and suitability of the GSV in poultry diets. The overall mean values for crude fibers (CF) (4.10±1.76) were high with wide range were associated with tannins and could suggest the limitation of using GSV in poultry diets. The overall mean values for ash content (3.51±1.52) was high and could be due to presence of contaminants among GSV from different sources. The overall mean values of the macro and micro minerals and the essential amino acids were generally low signifying the need for supplementation of minerals and amino acids in grain sorghum based diets for poultry diets. High mean values for condensed tannins (3.33%DM) as leucocyanidin equivalent and total phenolic compounds (25.0%DM) found in some of GSV could limit utilization of these grains in poultry diets. In the present study a low positive correlation was noted between color of the sorghum grains and nutrient composition, anti-nutritional components, and ME_n might suggest their similarity in nutritive values. The findings of the present study showed that Tanzanian GSV have high nutritive value and could partially replace maize in poultry feeding. However, their

full utilization in poultry diets requires a strategic improvement to reduce anticipated effects of noted anti-nutritional factors.

Keywords: *chemical composition, grain sorghum varieties, nutritive value, poultry feeds*

Introduction

In Tanzania, sorghum (*S. bicolor* (L.) Moench), (Mtama in the local Swahili language) is a second most widely grown cereal crop with a wide genetic diversity (Bucheyeki et al 2010). The crop is grown by majority of peasant farmers across the semi-arid areas in central regions of country with substantial annual production (Minde and Mbiha 1993). Sorghum is preferred because it is drought tolerant, grows well in marginal areas, and thus yields more than maize under harsh conditions. It is also used as a food security crop in rural areas (Roharbach and Kiriwaggulu 2007). Due to these attributes sorghum has a potential of being used as an alternative feed ingredient in commercial poultry feeds.

However, in Tanzania the use of grain sorghum in poultry feeds is not preferred by commercial livestock feed processors and farmers because it is viewed as inferior crop compared to maize and could affect poultry performance (Roharbach and Kiriwaggulu 2007). Information on nutritive value of the existing commercial grain sorghum varieties is not well documented in Tanzania. Thus, the main objective of the present study was to evaluate and document the nutritive potential of commercial sorghum varieties for poultry in Tanzania.

Materials and Methods

Study area

The study was carried out in purposively selected nine main grain markets located in urban and peri-urban areas of Singida, Dodoma and Morogoro regions, eastern central Tanzania. The regions are semi-arid with savannah climate characterized by short rainy period of four months between December and May with an average annual rainfall of 450mm; maximum temperature of 28°C and minimum temperature of 17°C, the climate favors sorghum production. The selected regions are the main grain sorghum producers, users and had pockets of sorghum-pearl millet groundnut based farming systems.

Identification of available grain sorghum varieties

A cross section survey in these regions was carried out to identify available commercial grain sorghum varieties (GSV) in the respective food markets. About 10 key informants in each respective market were selected and interviewed using a structured questionnaire so as to obtain qualitative and quantitative data on grain sorghum marketing profile for each identified grain GSV in the market. Most of the respondents were men who were either whole sellers or retailers of grain sorghum in the respective markets. All the types of grain sorghums sold by each interviewed whole-seller or retailer were recorded, identified based on physical characteristics and common local names. Thereafter identified varieties in markets were sampled and kept in dry paper bags for further verification and laboratory analysis.

Thereafter, the samples were ground through a 1 mm screen, packed in dry bottles, labeled and sent to the Department of Animal Science and Production (DASP) at Sokoine University of Agriculture (SUA) for the analyses of chemical composition and anti-nutritional factors.

Determination of Proximate, minerals and ME_n

Dry Matter (DM), Crude Protein (CP), Ether Extract (EE) and ash for the sorghum grain samples were determined using the AOAC (1990) standard methods. The minerals in grain sorghum samples were determined using atomic absorption spectro-photometric method described by AOAC (1990). All the analyses were done in duplicates. The values of calcium, phosphorus, magnesium and potassium were reported in percentage while sodium and iron were reported in mg/100g. Zinc, phosphorus, manganese and copper were reported in parts per million (ppm). The calorific (energy) value was calculated by a formula: ME (kcal/kg) = (21.98 x CP) + (54.75 x EE) + (35.18 x NFE) as expressed by NRC (1994) and expressed as MJ/kgDM.

Determination of anti-nutritional factors (condensed tannins, total phenol compounds and Phytic acids)

The condensed tannins in the ground samples of GSV were determined using Butanol/HCL method as described by Nitao et al (2001). The condensed tannins were determined as percent of leucocyanidin equivalent in accordance with the regression equation obtained from standard calibration plot. The equation expressed as $Y=0.005x-0.025$ (R^2 *=0.855).* Condensed tannins were expressed as mg / 100 mgDM. The total phenolic compounds in finely ground samples of grain sorghum were determined using the procedure described by Cioroi and Dumitriu (2009). Calculation of total phenols in the original sample was computed using the equation obtained by standard calibration plot. The equation expressed as $Y=0.001x-0.000$ (R^2 =0.997). Total phenols were expressed as g/100g. The phytic acid in finely ground sorghum samples were determined based on an iron to phosphorous ratio of 4:6 using the method as described by Mustafa et al (2003). Calculation of total Phytic acids in the original sample was computed by using the equation obtained by standard calibration plot. The equation expressed as $y = 1.5013x + 0.0441$ ($R^2 = 0.9528$). Phytic acids were expressed as μg /100gmDM.

Statistical analysis

All the obtained data were analyzed using descriptive statistic as described by Olawuyi (1996). The Statistical values that were calculated included the means and standard deviation.

Results

Commercial grain sorghum varieties

The results of commercial GSV sold in selected food markets are presented in Table 1. A total of twelve GSV were identified in this study. Seventy five percent (75%) of the GSV were landraces and the remaining (25%) were improved varieties. Fifty eight percent (58%) of GSV had white coat colour and the rest (42%) had red or yellow. Manyiang'ombe, Kakera, Mkombituna, Mankumba and Mwangurungi varieties were identified as being from Singida food markets and various districts, respectively. Mbangala variety was from Morogoro whereas Serena, Langalanga, Udo, Macia and Lugugu varieties were all sourced from various districts of Dodoma region. However, Serena and Tegemeo varieties were also found in Morogoro region markets.

Proximate composition, energy and sugar content and some amino acid profile

The results for proximate composition and energy content of grain sorghum are presented in Table 1. The study revealed high DM, CP, EE and ME_n values with a slight variation across grain sorghum varieties (GSV). Moreover, the CF values were high but with a wide (5.1%) range across GSV. The highest CF values were noted in Mwankurungi and lowest in Lugugu variety. In addition, the results showed high mean NFE content but with a wide (7.7%) range across GSV. The highest NFE values were observed in Mbangala and lowest in Mwankurungi variety. However, results showed a moderate ash values with a wide range amongst GSV. The findings for starch, sugar content and profile of some amino acids are presented in Table 2. High starch values and small proportions of sugar and amino acids were noted across GSV. The amount of lysine, tryptophan and methionine & cystine were small with insignificant variation across GSV.

Macro and micro minerals and Anti-nutritional factors

The results for macro and micro minerals are presented in Table 3. The results showed low values of calcium, phosphorus and magnesium amongst GSV. In addition, low levels of micro-minerals were noted across GSV. The results for anti-nutritional components of GSV are presented in Table 4. The study revealed high condensed tannin values with a wide variability across GSV. The lowest condensed tannins were observed in Tegemeo and highest in Mwankurungi variety. Further, results showed high total phenolic compound values with a wide range amongst GSV. Serena had highest level of total phenolic compounds and Lugugu variety had lowest relative to their counterparts. The values of phytates observed in this study were considerably low and did not differ much across GSV.

Table 1: Proximate composition and energy content of sorghum grains (%DM basis)

Variety	DM (%)	CF (%)	EE (%)	CP (%)	Ash (%)	NFE (%)	ME MJ/Kg DM
Manyiangombe	88.3	3.7003	3.29	11.3	2.44	79.3	13.5
Kakera	89.2	2.83	4.0508	10.44	5.65	77.03	13.2
Serena	87.3	6.55	2.66	12.7	2.45	75.6	12.9
Tegemeo	88.7	3.62	2.71	11.09	1.79	80.79	13.5
Langalanga	88.3	2.59	3.41	11.04	2.28	80.68	13.7
Mwankurungi	88.1	6.84	2.81	11.4	4.53	74.5	12.7
Mkombituna	88.3	3.83	2.84	10.69	4.52	75.3	12.7
Udo	88.3	5.61	2.88	10.78	2.66	78.1	13.1
Mankumba	88.07	6.056	3.12	11.3	4.95	74.6	12.7
Mbangala	88.1	1.98	2.76	10.84	2.206	82.4	13.8
Lugugu	88.3	1.74	3.35	10.46	6.43	78.4	13.3
Macia	88.4	3.79	3.88	11.6	2.67	78.3	13.4
Mean	88.3	4.095	3.15	11.09	3.55	77.92	13.2
Maximum	89.2	6.84	4.0508	12.7	6.43	82.4	13.8
Minimum	87.3	1.74	2.66	10.44	1.79	74.6	12.7
±SD	0.437	1.76	0.462	0.626	1.57	2.59	0.391

DM: Dry *Matter; CP: Crude Protein; CF: Crude Fiber: EE: Ether Extract: NFE: Nitrogen Free Extract: ME: Metabolizable Energy; ME (kcal/kg) = (21.98x CP) + (54.75 x EE) + (35.18 x NFE) (Janssen, 1989).*

Table 2::Starch, sugar and some essential amino acids of sorghum grains (%DM basis)

Variety	Starch	Sugar	Lysine	Tryptophan	Methionine+cystine
Manyiangombe	70.58	1.22	0.0734	0.885	3.45
Kakera	72.4	1.14	0.0362	0.954	2.88
Serena	66.8	0.227	0.0191	0.788	2.97
Tegemeo	68.4	1.58	0.000799	0.923	2.99
Langalanga	73.4	0.0691	0.0002037	0.695	2.19
Mwankurungi	65.9	0.0176	0.0436	0.625	2.27
Mkombituna	72.3	0.867	0.0577	0.662	2.46
Udo	67.2	0.0985	0.004921	0.486	1.86
Mankumba	68.6	0.0887	0.0722	0.624	2.085
Mbangala	76.1	1.081	0.00493	0.926	3.56
Lugugu	73.7	1.099	0.0748	0.717	2.53
Macia	68.5	1.601	0.0163	1.016	3.021
Mean	70.32	0.744	0.0664	0.752	2.69
Maximum	76.1	1.601	0.0734	1.016	3.56
Minimum	65.9	0.0176	0.0002037	0.486	1.86
±SD	3.22	0.634	0.1201	0.154	0.54006

Table 3: The macro and micro mineral profile of Tanzanian grain sorghum varieties (% DM basis)

Variety	Ca (%)	P (%),	Mg (%)	K (%)	Fe (mg/100 gDM)	Na (mg/100gDM)	Zn (PPM)	Cu (PPM)	Mn (PPM)
Manyiang'ombe	0.00984	0.305	0.177	0.366	32.1	7.96	8.76	1.98	18.9
Kakera	0.0114	0.302	0.205	0.353	44.5	4. 98	8.68	3.12	29.9
Serena	0.0113	0.382	0.2051	0.464	21.6	6.69	8.18	1.19	19.1
Tegemeo	0.00938	0.328	0.171	0.407	5.50	6.13	7.05	1.17	11.3
Langalanga	0.0119	0.358	0.214	0.383	56.8	7.51	9.78	1.18	18.9
Mwankurungi	0.0124	0.276	0.166	0.393	64.0	10.3	8.11	1.18	18.9
Mkombituna	0.00942	0.276	0.169	0.399	182	3.68	9.11	1.18	15.1
Udo	0.00984	0.343	0.172	0.399	18.2	4.35	6.41	1.18	18.9
Mankumba	0.0248	0. 314	0.192	0.4604	93.6	10.3	9.14	3.16	34.1
Mbangala	0.00476	0.376	0.199	0.359	23.3	5.95	7.11	1.18	18.9
Lugugu	0.0115	0.276	0.169	0.331	96.3	8.42	9.45	5.12	30.2
Macia	0.00772	0.378	0.178	0.4061	14.4	5.71	8.41	1.18	18.8
Mean	0.0115	0.326	0.185	0.393	54.4	6.87	8.35	1.90	21.1
Maximum	0.0248	0.382	0.214	0.464	182	10.5	9.14	5.12	30.2
Minimum	0.00476	0.276	0.166	0.331	5.50	3.68	6.41	1.17	11.3

Table 4: The Anti-nutritional factor contents in grain sorghum varieties (%DM basis)

Variety	Type	Coat colour	DM (%)	Tannin (% DM)	Total Phenols (% DM)	Phytic acids (µg/100gDM)
Manyiang'ombe	Landrace	Red	88.3	2.61	26.9	1.56
Kakera	Landrace	White	89.2	2.20	5.23	0.76
Serena	Improved	Red	87.3	2.78	52.5	4.23
Tegemeo	Improved	White	88.7	2.18	10.6	1.44
Langalanga	Landrace	White	88.3	2.53	6.13	3.69
Mwankurungi	Landrace	White	88.0	5.76	32.0	0.33
Mkombituna	Landrace	Red	88.3	3.80	12.9	1.22
Udo	Landrace	Red	87.3	3.33	45.1	6.98
Mankumba	Landrace	White	89.0	5.20	34.7	3.44
Mbangala	Landrace	White	88.1	2.60	5.55	1.25
Lugugu	Landrace	White	88.3	3.18	2.44	2.25
Macia	Improved	White	88.4	3.76	10.1	1.93
Mean				3.33	22.2.	2.42
Maximum				5.76	52.5	6.98
Minimum				2.18	2.44	0.33
SD±				1.14	20.0	1.87
CV				2.80	4.11	5.99
R^2				1.00	1.00	1.00

Discussion

The high DM content observed in the present study across GSV indicates low in moisture content. These findings suggest Tanzanian GSV could produce high valuable feed component that could be conducive for storage of poultry feeds. DM content below 85% leads to deterioration of feed ingredients and feed products due to growth of molds and fungi particularly in tropical countries (Hamito 2010). These observations agree with Goromela (2009) and Mutayoba et al (2011).

In addition, the high CP content with a slight variability amongst GSV may indicate the GSV are grown in similar climatic and agronomic conditions such rainfall, temperature, soil fertility and time of harvest (Bryden et al 2009). These results suggest the use of studied GSV in poultry diets could probably contribute to high proportion of dietary CP and reduce the cost of supplementing with high protein ingredients in poultry diets. However, the CP quality depends on the availability of essential amino acids and their availability to the animal. In the present study low levels of amino acids such as lysine, methionine& cystine and tryptophan indicate poor quality proteins in studied GSV. These findings demonstrate that lysine and methionine are amongst the most limiting amino acids in GSV.Thus, the utilization of studied GSV in poultry diets may require considerable use of high quality protein such as fish meal or soybeans to curb the effect of limiting amino acids. These results are congruent with the research report of Gualtieri and Rapaccini (1990) and Ebadi et al (2005).

Moreover, the moderate fat (EE) content with slight differences observed in the present study amongst studied GSV demonstrate that the GSV probably grown in similar growing conditions, soil type or are harvested within the same period of time. These findings suggest the use of studied GSV in poultry diets could relatively improve the feed keeping quality and shelf life and produce body fat and products which may pose little health risks associated with cholesterol to humans compared to maize. Unsaturated fats from cereals such as linoleic and oleic become rancid quickly and produce soft fat body fat in poultry (MacDonald et al 1999) and may influence low density lipoprotein (bad cholesterol). These results concur with results observed by Mutayoba (2011).

The ash content observed across grain sorghum varieties in the present study were within the levels reported by other authors (Kriegshauser et al 2006 and Mutayoba et al 2011). These findings indicated a wide range (1.7 to 6.9%DM) of ash content across grain sorghum varieties. The wide range of ash in grain sorghum may be attributed by various factors such as variability in soil type, storage condition, climate and contaminants of inorganic substances such as sand (Bryden et al 2009). The ash represents inorganic constituents of feed ingredients but contain other materials from the contamination of the feed ingredients. The ash levels in the present study was not reflected the amount of calcium, phosphorous and other minerals, thus could be an indication of contamination by soil or sand particles during harvesting and or processing. These results suggest the use of GSV in poultry diets could require appropriate harvesting and processing methods to avoid contamination.

The CF is used to define a variety of indigestible polysaccharides including cellulose, hemicelluloses, pectins, oligosaccharides, gums and various lignified compounds. The high CF content with a wide range across GSV observed in the present study could indicate variability of tannin content in GSV. These findings could reflect high and variability of tannin content in GSV (Dykes and Rooney 2006). The GSV with highest CF also had highest condensed tannins in the present study. The contribution of polyphenols to the lignin fraction of the GSV fibre could be responsible for the higher values of fibre in the high-tannin variety. The CF value is important in major feed ingredients in livestock diets. High CF values in feed ingredients tend to increase dietary CF values that could impair digestibility in mono-gastric animals. Further, high dietary fibre has certain adverse effects on the availability of some nutrients. These results suggest the use of studied GSV in poultry diets could negatively affect digestibility and ultimately performance. Thus, the utilization of studied GSV in poultry diets requires a strategy which could reduce tannin content and eventually lower CF content.

The high ME_n with a narrow range observed in this study across GSV agree with observations by Donkoh and Attoh- Kotoku (2009). These results indicate that the GSV are either grown in similar climate or agronomical conditions. Energy in GSV content depends on the availability of starch. However, the availability of starch is influenced by the interaction between proteins, cell walls, non-starch polysaccharides, kafirins and tannins (Chandrasheker and Kirleis, 1988; Kavitha and Chandrasheker, 1997 and Duodu et al 2003). All these factors are either genetically or environmentally controlled. These findings suggest the high ability of studied GSV to supply substantial energy in poultry diets.

The phenolic compounds may include simple phenols, phenolic acids, flavonoids, hydrolysable tannins, condensed tannins and lignin (Hahn et al 1984).The high total phenolic compounds with a wide range across GSV observed in the present study might indicate high genetic differences amongst GSV. These findings suggest the utilization of studied GSV in poultry diets could produce stable concentrates (Dykes and Rooney, 2006). However, high total phenolic compounds may limit the use of GSV in poultry feeds due to the presence of condensed tannins and lignin which impair utilization of nutrients and eventually could negatively affect performance in poultry. Moreover, the high content of condensed tannins with a wide range across GSV might indicate that the GSV are genetically different (Etuk, 2008). These results indicate that the studied GSV belong to type III containing high tannins content (Dykes and Rooney 2006). The high condensed tannins might limit their use in poultry diets. Tannins are polyphenolic compounds, which have ability to form complexes with metal ions and with macro-molecules such as proteins and polysaccharide and eventually impair digestion and make them unavailable to poultry (Dei et al 2007). Dietary tannins reduce feed efficiency and weight gain in chicks (Armstrong et al 1994). However, tannins are tolerable up to 1% in poultry diets (Ravindran et al 2006). Thus, the use of studied GSV in poultry diets might require a strategic improvement to reduce negative effects of tannins such as chemical and physical treatment of GSV or supplementation of nutrients bound or denatured by tannins (Gualtieri and Rapaccini 2009). These observations were consistent with the results reported by Tabosa et al (1995) and Medugu et al (2010).

The low of phytates observed across GSV in the present study conflict with other research reports (Wanisca and Rooney 2000; Mustafa et al 2003). The reasons for these results are not clear but could indicate GSV are grown in dry climatic conditions without phosphorus fertilizer applications (Bassiri and Nahapetion 1977 and Asada et al1969). The low phytic content observed in this study across GSV could suggest non-Phytic phosphorus and availability minerals. There is a negative correlation between phytates content and bioavailability of phosphorus (Ahmad and Sattar 2003). About 30-90% of total phosphorous is form of phytates (MacDonald et al 1999). Moreover, phytic acid is not desirable dietary agent because it chelates multi-valent metal ions, particularly phosphorus, zinc, calcium and iron and proteins which results in insoluble salts leading to unavailability of those minerals (Bryden et al 2007). These results suggest the utilization of GSV in poultry diets could minimize supplementation of minerals particularly phosphorous and calcium or use of Phytase to curb the anti-nutritional effect of phytates in poultry.

Conclusions

It can be concluded that Tanzanian GSV have high nutritive value and quality and could partially replace maize in poultry feeding. But supplementation of mineral and amino acids is essential to optimize their nutritive value for poultry. However, their full utilization in poultry diets could require a strategic improvement to reduce effects of anti-nutritional factors to improve their feeding value.

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CHAPTER FOUR

4.0 PAPER II

Effects of *Moringa oleifera* leaf meal and molasses as additives in grain sorghum based diets on performance of growing chicks in Tanzania.

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Relative effects of *Moringa oleifera* leaf meal and molasses as additives in grain sorghum based diets on performance of growing chicks in Tanzania

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Abstract

The effects of supplementing *Moringa oleifera* leaf meal (MOLM) and molasses to grain sorghum based diets on performance of growing chicks was studied in a 2x3 factorial experiment for 8 weeks. The study used two grain sorghum varieties: high (HTS) and low (LTS) tannin; three supplementary strategies of non-supplemented, 10% DM- molasses and 10% DM-MOLM to make six diets, CTL_1, SCM_2, MOL_1 and CTL_2, SCM_2 and MOL_2 respectively.

Survival, colour of body parts, growth parameters and feed conversion ratio were not influenced by type of sorghum. The DMI was higher in HTS and leg abnormality incidence (LAI) was higher in LTS. Survival and growth parameters were not influenced by type of supplementation. The supplementation of molasses aggravated, while supplementation of MOLM prevented, occurrence of LAI. The birds fed molasses showed paleness but those fed MOLM showed yellowing of body parts. The DMI and FCR were better in birds fed CTL_1 compared with fed CTL_2 based diets. Supplementation of molasses and MOLM improved DMI and FCR in HTS compared with LTS. HTS based diets in growing chicks should be supplemented with energy sources, minerals and pigments; while LTS based diets should be supplemented with minerals and pigments. Sugar cane molasses improves FCR in HTS but requires supplementation of minerals and pigments. Use of molasses as an additive to LTS based could impair FCR and aggravate LAI and is not recommended. The use of MOLM in HTS and LTS based diets can improve FCR and prevent prevalence of LAI and has the potential to produce coloured products.

Key words: *agro forestry, leg weakness, minerals, mortality, pigments*

Introduction

Worldwide sorghum (*Sorghum bicolor*) is the second cereal crop used after maize in poultry feeds (FAO 2005). Sorghum plant crop is preferred because it has adaptive features that favor its growth in areas where other cereals cannot survive (Haussmann et al 2002; Pray and Nagarajan 2009). The nutritive value of grain sorghum is well established and is comparable to maize in poultry feeding (BSTID-NRC 1996 and Ebadi et al 2005). However, the presence of anti-nutritional factors such as condensed tannins has been compromising their feeding value. Tannins form complexes which may cause unavailability of nutrients and may impair feed palatability (Awad et al 2001). In addition, tannin levels of more than 1% in poultry diets reduce dry matter intake, body weight gain and feed efficiency (Hassan et al 2003 and Ravindran et al 2006).This situation has prompted scientists to devise various strategies for improving the feeding value of high tannin grain sorghum (HTS) in poultry. Various strategies and techniques aiming at reducing tannin content, improving digestibility or supplementing unavailable nutrients are well documented or suggested (Cramer et al 2003 and Ravindran et al 2006). However, the adoption of proposed strategies and techniques for improvement of grain sorghum feeding value have not been forthcoming since some of them are expensive, cumbersome or not practical under farmer conditions in developing countries. Thus, supplementing the deficient nutrients in grain sorghum based diets using cheap and locally available feed ingredients could be a viable strategic option in improving feeding value of grain sorghum in poultry.

In the Tanzanian context, *Moringa oleifera* leaf meal (MOLM) and final sugar cane molasses could be used as feasible feed additives in improving the feeding value of grain sorghum for poultry. *Moringa oleifera* is well distributed, survives in harsh conditions and contains high annual dry matter production as well as balanced amino acids compared to soybeans (Makkar and Becker 1996; Makkar and Becker 1997 and Becker and Makkar 2000). Furthermore, molasses by-product in sugar manufacturing industries is rich in highly digestible carbohydrates and minerals, exerts a tonic effect, induces palatability and could eliminate dustiness in poultry feeds. Also, the utilization of MOLM and molasses in poultry feeding is well documented (Keshavarz et al 1980; Kakengi et al 2007 and Katanga 2013). However, there is scanty scientific evidence showing the effectiveness of MOLM and molasses in improving the feeding value of grain sorghum. Therefore, based on this gap, a comparative study was conducted to evaluate the effects of supplementing MOLM and molasses to grain sorghum variety based diets on performance of growing chicks. It was hypothesized that MOLM and molasses supplementation to HTS and low tannin grain sorghum (LTS) based diets could equally improve the feeding value of grain sorghum and ultimately improve performance of growing chicks.

Materials and Methods

Location of the study

This study was conducted at poultry unit in Department of Animal Science and Production (DASP) of Sokoine University of Agriculture (SUA). It is located between 6 ^{o} and 7 ^{o} South and 37 ^{o} and 38 ^{o} east within an altitude of about 500 to 600m above sea level at the foot of Uluguru plateau mountains within Morogoro Municipality in Eastern part of Tanzania. It is characterized by ambient temperature between 20-27 °C in the coolest months of April to August and 30 - 35 °C during the hottest month of October to January. The annual rainfall ranges from 600-1000mm.

Experimental feed ingredients and diets

The two grain sorghum varieties (GSV) for HTS and LTS used in the present study were locally known as Serena and Mbangala respectively. The GSV were purchased from local food market, ground with a hammer mill and passed through 2mm sieve to produce grain sorghum meal and stored in clean nylon bags for the entire study period. MOLM was obtained by harvesting *Moringa oleifera* leaves from DASP compound. Moringa tree branches were cut, spread out and dried under the shade for a period of 3 to 4 days, threshed carefully before milling with hammer mill to produce MOLM and stored in nylon bags for the entire study period. Final sugar cane molasses was purchased from Mtibwa sugar processing company, packed and stored in plastic containers. Other feed ingredients used in this study were purchased from local livestock feed dealers. The proximate composition, ash, phosphorus and calcium for feed ingredients and diets were determined according to the methods of AOAC (1990). Metabolizable Energy (ME/ kcal/kg) content of feed ingredients and experimental diets were estimated by prediction equations as expressed in NRC (1994) and MOLM, ME/Kgcal was estimated by prediction equation established by Carpenter and Clegg (1956). Condensed tannins in GSV and MOLM were determined using butanol/HCL method. The diets were formulated to meet or exceed the MacDonald (1998) nutrient recommendations for growing chicks.

Experimental design and dietary treatments

Two hundred and seventy (270) day old commercial hybrid layer strain chicks with a mean weight of 40.9 gm were randomly allotted to six dietary treatments arranged in three replicates in a 2x3 factorial design for HTS (4.72% DM tannin) vs LTS (2.32%DM-tannin) and three supplement strategies (SPS): non- supplemented (control), 10% DM - molasses and 10% DM - MOLM designated as CTL_1, SCM_1 and MOL_1 and CTL_2, SCM_2 and MOL_2 respectively. The diets (D_1-D_6) are shown in Table 1.

Experimental birds and their management

Birds were wing banded and randomly allocated to the six dietary treatments in three replicates per treatment. Each dietary treatment had 45 birds and 15 birds per replicate. Birds in each replicate were randomly placed in a single pen in an open ended poultry house in a deep litter system. Birds were weighed individually immediately after arrival using electronic sensitive weighing machine to obtain individual initial bird weight and thereafter on a weekly basis for body weight changes. Birds were vaccinated against Newcastle and Gumboro disease and subjected to a preliminary period of one week so as to acclimatize them to the experimental pens and diets. Water and feed were provided without restriction. Light was supplied by electric bulbs and sun at night and day time, respectively. Feed offered and leftovers were recorded daily. The amount of feed offered was 20 percent above the expected requirement so as to allow liberal consumption. The residual feed was weighed daily before supplying another feed for the next day. Feed intake was calculated by taking the difference between the feed offered and residual in each replicate. Daily weight gain (DWG) was calculated as individual average weight gain over a period of time. DFI (g/bird) over DWG (g/bird) was used to calculate FCR. Birds with abnormal legs were recorded daily. Then leg abnormality incidences (LAI) were calculated as the ratio of birds with leg abnormalities to total number of birds available x 100.

Statistical analysis

Data on growth, DMI, FCR and LAI parameters were analyzed in accordance with a 2x3 factorial design, using the General Linear Model procedure of SAS software version 9.1 for windows (2007). The Least Square Difference was used to compare means of each variable. The analytical model for growth parameters, DMI and FCR was as follows:

$y_{ijk} = \mu + V_i + S_j + (VS)_{ij} + b(X_{ijk} - x) + E_{ijk}$

Where:

Y_{ijk} = Observation of k^{th} bird assigned to i^{th} GSV subjected to $_{jth}$ SPS;

μ = overall mean to all observations; V_i = effect of GSV based diet (HTS or LTS);

S_j = the effect of SPS based diet (CTL, SCM or MOL);

$(VS)_{ik}$ = the interaction between GSV and SPS;

X_{ijk} = Initial weight of an individual bird;

x = Overall mean for initial body weight;

b = regression coefficient of Y_{ijk} on X_{ijk};

e_{ijk} = random error

Table 1: Gross composition of experimental chick diets

Ingredients	CTL_1	CTL_2	SCM_1	SCM_2	MOL_1	MOL_2
Moringa leaf meal	-	-	-	-	11.3	11.3
Molasses	-	-	14.4	14.4	-	-
Sorghum meal (High Tannin)	62.5	-	61.4	-	61.4	-
Sorghum meal (Low Tannin)	-	62.3	-	61.4	-	61.4
Fish meal	16.0	16.0	16.0	16.0	16.0	16.0
Sunflower seed meal	16.7	16.9	16.9	16.9	16.9	16.9
Limestone	2.00	2.00	2.00	2.00	2.20	2.20
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Bone meal	2.00	2.00	2.00	2.00	2.00	2.00
Salt	0.40	0.40	0.40	0.40	0.40	0.40
Chemical composition, %						
Dry matter	89.6	89.5	87.1	88.3	89.0	89.4
Crude protein	23.6	24.5	21.6	22.5	26.1	26.4
Crude fiber	9.60	9.27	7.41	6.50	10.6	8.03
Ether extract	7.66	8.46	5.97	6.76	7.38	6.87
NFE	47.1	47.2	51.2	52.2	39.7	39.0
Lysine	1.00	1.11	1.03	1.38	1.40	1.54
Methionine + cystine	0.86	0.67	0.70	0.77	0.75	0.82
Tryptophan	0.23	0.28	0.29	0.31	0.26	0.28
Tannin	2.60	1.30	2.15	1.05	2.55	1.36
ME (MJ/kgDM)	10.8	11.1	12.05	12.4	11.7	12.0

Results

General condition and performance of birds fed GSV diets

No mortalities were observed for the entire experimental period. Paleness of body parts were observed in HTS and LTS dietary groups. The DMI was higher by 21.9% in HTS compared to LTS dietary groups. The LAI characterized by swelling of hock joints and slipped tendons were noted in 8.5% of experimental birds. However, LAI was higher by 436% in LTS compared to HTS dietary group.

General condition and performance of birds fed SPS diets

During the study no mortality cases were recorded amongst SPS dietary groups for the entire study period. Paleness of body parts was observed in CTL and SCM dietary group. But, yellowing of body parts was noted in MOL dietary group. The LAI was higher in SCM by 12.3% compared to CTL dietary group. However, there was no LAI observed in MOL dietary group. The growth parameters did not differ among the SPS dietary groups. However, there was a significant interaction between sorghum varieties and SPS on DMI and FCR. The DMI decreased in SCM_1 and MOL_1 by 10.4% and 14.9% respectively whilst it increased in SCM_2 and MOL_2 by 37.5% and 10.7% respectively. The FCR was improved in SCM_1 and MOL_1 by 7.6% and 11.2% respectively whilst it was worse in SCM_2 and MOL_2 by 44.6% and 20.8%, respectively.

Table 2: Performance parameters of growing chicks fed high and low tannin grain sorghum variety based diets (0-8 weeks of age)

	HTS	LTS	SEM	Prob
Initial weight, g	43.2^{a}	40.1^{b}	0.81	0.001
Weight gain, g/d	11.8	11.9	0.21	0.543
DM intake, g/d	42.3^{a}	34.7^{b}	1.59	0.015
Feed conversion ratio, g feed/g gain	3.93	3.39	0.13	0.086
Leg abnormality incidences, %	2.22^{b}	11.9^{a}	2.83	0.037

Table 3: Performance parameters of growing chicks fed Moringa (MOL) and molasses supplemented (SCM) sorghum based diets (0-8 weeks of age)

Parameter	Supplementary diet				
	CTL	SCM	MOL	SEM	Prob
Initial weight, g	39.7^{b}	38.5^{b}	46.9^{a}	2.42	0.002
Final weight, g	668	664	593	4.10	0.421
Daily weight gain, g	12.1	11.6	11.6	0.545	0.276
LAI, %	8.88^{ab}	12.3^{a}	0.00^{c}	8.48	0.078

^{ab} Means along the same row with different superscripts differ at $p<0.05$)

Table 4: Interaction of tannin levels in grain sorghum variety and type of supplementary strategy on dry matter intake and feed conversion ratio of growing chicks (0-8 weeks of age).

Parameter	HTS			LTS			SEM	p
	CTL_1	SCM_1	MOL_1	CTL_2	SCM_2	MOL_2		
DM intake, g	46.2^{a}	41.4^{ab}	39.3^{ab}	29.9^{b}	41.1^{ab}	33.1^{ab}	4.10	0.321
Feed conversion, g feed/g gain	4.19^{a}	3.87^{ab}	3.72^{ab}	2.79^{b}	4.035^{ab}	3.37^{ab}	0.444	0.338

^{ab} Means along the same row within main effects with different superscripts differ at $p < 0.05$

Discussion

The absence of bird mortality observed in the present study amongst HTS and LTS dietary groups indicates that tannin had no lethal effects on survival. Dietary tannin levels more than 3% have detrimental effects on survival according to Ali and Mahmood (2013). These results agree with those of previous studies (Hassan et al 2003).

Paleness of body parts (shank, feet and beak) observed in HTS and LTS dietary groups are attributed to the absence of xanthophylls and carotenoid pigments in sorghum grain (NRC 1994; Sauvant et al 2004). These results concur with those of other research results (Moura et al 2011; Garcia et al 2013). Meat and egg yolk colour is a valuable quality attribute evaluated by consumers. Colored poultry products are an indication of meat freshness and directly influence consumer's purchase decision. The success of a product depends on consumers' acceptance and its quality and appearance are amongst the most valuable features. Therefore utilization of sorghum-based diets in poultry requires addition of synthetic or natural pigments to improve coloration of the products (Assuena et al 2008; Garcia et al 2013; Schiedt 1998).

The prevalence of LAI observed amongst the chickens fed sorghum grain is congruent with those of other studies (Jacob et al 1996; Hassan et al 2003). These results may be attributed to inadequacy of minerals particularly phosphorus and calcium in sorghum grain and negative effects of tannins or phytates (Khalid et al 2003; Sebastian et al 1998). Multiple phenolic hydroxyl groups of tannins may form stable complexes with metal ions and other macromolecules like polysaccharides (Kondo et al 2007) and interact with minerals to form precipitates and thus reduce their availability (Hassan et al 2003). The minerals which are ionized in the stomach (iron, calcium, magnesium, sodium and potassium) are prone to a variety of absorption interferences (Arigator and Samman 1994). Tannin-nutrient interaction may be one of the means by which tannins affect the digestive processes resulting in reduced availability of the nutrients in the gut. Moreover, phytic acid is not a desirable dietary agent because it chelates multi-valent metal ions, particularly phosphorus, zinc, calcium and iron and proteins which results in insoluble salts leading to unavailability of the minerals (Bryden et al 2007). Calcium and phosphorus are the most abundant minerals in bone. Therefore, deficiency of calcium and phosphorus could negatively affect development of bone matrix and exert bone deformities. Bone is a highly complex structure, and its composition varies according to nutritional status of the animal (Musharaf and Latshaw 1991; Sebastian et al 1998). The results suggest the use of sorghum based diets in growing chicks should be accompanied with high supplementation of minerals or use of phytase to curb the prevalence of LAI. The higher LAI in LTS compared with the HTS dietary group possibly indicates presence of more phytates in LTS.

The higher DMI observed in HTS than LTS dietary groups corroborates with data from other researchers (Nyakoti and Atkinson 1995; Nyakoti et al 1997). This finding indicates energy inadequacy in HTS and energy adequacy in LTS. The lower Metabolizable energy in HTS due to negative effect of tannins (Elkin et al 1996) may be the reason for the increase of DMI as compensatory effect. These findings suggest HTS should either be supplemented with other energy sources or treated to reduce the amount of tannins.

The comparable growth performance observed amongst HTS and LTS dietary groups was attributed to the increase of DMI observed in HTS dietary group which perhaps offset the negative effect of tannins on nutrients such as proteins and amino acids responsible for body accretion. These findings suggest utilization of HTS in diets of growing chicks has no detrimental effects on growth performance. These results concur with those of other previous studies (Ambula et al 2001; Gadzirayi et al 2012).

The moderate lower FCR values observed in HTS than LTS dietary groups though were not significant suggest that the use of HTS in growing chicks could impair FCR. These findings were attributed by higher DMI noted in HTS compared to LTS dietary group. These findings suggest use of HTS in growing chicks should be supplemented with cheap energy sources to curb negative effects of tannins and reduce DMI. These results are in agreement with those of other research reports (Sannamani 2002).

The paleness of body parts (shank, feet and legs) observed amongst SCM dietary groups could be attributed to the absence of xanthophylls and carotenoid pigments in molasses. Yellowing of body parts noted in moringa leaf meal diets compares well with those of other research reports (Kaijage 2003; Olugbemi 2009). These findings may be attributed to the presence of xanthophylls and carotenoid pigments in moringa leaf meal. The leaves of moringa are rich in biologically active xanthophylls, the carotenoid which causes yellowing of skin and shank. Therefore utilization of moringa leaf meal as additive in sorghum based diets could improve colour of poultry products and reduce the expense of using synthetic pigments.

The higher LAI in SCM compared to CTL dietary group may be attributed to the effect of sugar cane molasses (Alvarez 1977; Ly 1990) which probably affects the bioavailability and utilization of phosphorous and calcium. Nutrient losses caused by final molasses may be partly due to osmotic effects caused by the great quantity of potassium ions or the laxative effect associated with insufficient intestinal saccharase used for complete hydrolization of sucrose (Obando et al 1969; Ly and Velázquez 1969). These findings suggest that 10% supplementation of final sugar molasses to sorghum-based diets could exaggerate LAI in growing chicks.

The interaction of GSV and SPS on DMI and FCR observed in the present study indicates energy adequacy in LTS and inadequacy in HTS due to the tannin effect. The decrease of DMI and FCR in SCM_1 and MOL_1 compared to CTL_1 dietary group was attributed to the effect of molasses and moringa leaf meal compensating for deficiency in energy in HTS. The increase in dietary energy concentration tends to decrease DMI in poultry (MacDonald et al 1998; NRC 1994).Therefore, these findings suggest utilization of molasses and MOLM as an additive to HTS based diet in growing chicks could improve DMI and eventually FCR.

However, the increase of DMI and FCR observed in SCM_2 compared to CTL_2 dietary group is not clear but may be attributed to the high palatability of molasses or the negative effect of molasses on energy utilization (Alvarez 1977 and Ly 1990). Thus, use of sugar cane molasses as an additive to LTS is not beneficial as it could partially aggravate DMI and ultimately impair FCR. Similarly, the increase of DMI and FCR observed in MOL_2 compared to CTL_2 dietary groups may be associated with either bulkiness or high palatability of MOLM (Kaijage 2003; Makanjuola et al 2014).The utilization of MOLM as an additive to LTS could partially increase DMI and impair FCR.

Conclusions

- It is possible to utilize high tannin sorghum as the main source of energy in commercial growing chicks without detrimental effects on growth and mortality of birds.
- High tannin sorghum should be accompanied by supplementation of cheap energy sources to improve feed conversion ratio, by minerals to prevent prevalence of LAI and lack of pigments to enable production of coloured poultry products desired by the consumers.
- Moringa leaf meal could be the most appropriate additive to sorghum based diets for growing chickens as its utilization could minimize expenses of using synthetic minerals and pigments.

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CHAPTER FIVE

5.0 PAPER III

Effects of *Moringa oleifera* leaf meal and molasses additives in grain sorghum based diets on haematology and serum bio-metabolites for growing chicks.

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Effects of *Moringa oleifera* leaf meal and molasses additives in grain sorghum based diets on haematology and serum bio-metabolites for growing chicks.

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Abstract

The effects of adding *Moringa oleifera* leaf meal (MOLM) and final sugar cane molasses (SCM) to grain sorghum based diets on hematological parameters and serum bio-metabolic indices of growing chicks were studied in a 2x3 factorial experiment after 8th week of age. The study used two grain sorghum varieties (GSV): high (HTS) and low (LTS) tannin; three supplementary strategies for non-supplemented GSV(control), 10% DM- SCM and 10% DM-MOLM supplemented GSV, to make six dietary treatments. The lymphocytes and oesinophils ratios and red blood cells (RBC) were higher whilst monocyte ratio was lower in HTS compared to LTS. The interaction between type of GSV and type of SPS was noted on lymphocyte, oesinophils and monocyte ratios. The addition of SCM and MOLM to HTS based diets (CTL_1) decreased lymphocyte and oesinophil ratios whilst increased monocyte ratios. But, addition of SCM to LTS based diets decreased lymphocyte whilst increased monocyte. Addition of SCM increased total serum protein value. It can be concluded that use of HTS lead to disruption of the immune system and nutritional stress in growing chicks. The addition of SCM and MOLM to HTS diets alleviate the nutritional stress and improve humeral immunity of growing chicks whereas addition of SCM to LTS diet lead to nutritional stress and disrupt immune system. Therefore, addition of 10%DM- MOLM and 10%DM- SCM in HTS based diets is suggested but addition of SCM in LTS diets is not recommended.

Key words: Growing chicks, grain sorghum, sugar cane molasses, moringa leaf meal, immune system

Introduction

The energy portion represents the largest dietary ingredient in poultry diets. Maize has been the major source of energy in poultry in most tropical countries. However, maize has been scarce and expensive due to a decreasing trend in production as a result of climate change and stiff competition between human and livestock species. This situation could be improved by using the alternative energy sources which are locally adaptive, cheap, less competitive and available. The grain sorghum is a potential alternative to maize in poultry feeding. It is adaptive to harsh conditions and produced extensively in the tropical semi-arid areas (Abdulla and Gamar, 2011). The role of grain sorghum in poultry feeding is well documented (Parthasarathy *et al.* 2005; Travis *et al.* 2006 and Issa *et al.* 2007). However, most of the sorghum varieties grown in tropics contain high tannin concentration which limits their utilization in poultry feeding. Nevertheless, in tropics cultivation of high tannin grain sorghum varieties is preferred due to agronomical and human health uses (Awika and

Rooney, 2004). The negative effects of tannins on hematological and serum metabolic parameters have been reported in poultry (Medugu *et al.* 2010). The negative effect of tannin on metabolic and physiological functions of tissues and organs in animals is reflected in serum biochemical parameters (Mellesse *et al.* 2011b). The toxicity, pathological status, abnormality and malfunctions of animals due tannins may be reflected through hematological parameters (Josh *et al.* 2002). This situation has prompted researchers to look for better options that could limit unfavorable effects of tannin grain sorghum and eventually improve health status of birds.

In Tanzania, MOLM and final sugar cane molasses seem to be the best options due to being cheap, locally available, nutritious and low in anti-nutritional factors (Anwar et al, 2007). MOLM is rich in protein and amino acids (Marker and Becker, 1997; Melesse, 2011). Molasses is rich in energy content. Currently, there is scanty information specifying the effectiveness of MOLM or molasses in alleviating the adverse effect of tannins on metabolic and physiological functions of tissues and organs or and body functions and healthy in poultry. Therefore, based on this gap, a study was conducted to evaluate the effects of supplementing MOLM and molasses to grain sorghum variety based diets on hematological parameters and serum biochemical indices. It is hypothesized that supplementation MOLM and molasses to HTS could improve the hematological parameters and serum biochemical metabolic indices and subsequently healthy of growing chicks.

Materials and Methods

Location of the study

This study was conducted at the poultry unit of the Department of Animal Science and Production (DASP) in the Sokoine University of Agriculture (SUA), located between 6° and 7 ° South and 37 ° and 38 ° east and at an altitude of about 500 to 600m above sea level at the foot of Uluguru Plateau Mountains within Morogoro Municipality in Eastern part of Tanzania. It is characterized by ambient temperature between 20-27 °C in the coolest months of April to August and 30 - 35 °C during the hottest month of October to January. The annual rainfall ranges from 600 to1000mm.

Experimental procedure

Two hundred and seventy (270) day old commercial hybrid bovan layer strain chicks were randomly allotted to three replicates in six dietary treatments arranged in a 2x3 factorial experiment for two grain sorghum varieties (GSV) with high and low tannin values and three supplementary strategies (SPS). The diets consisted of GSV for high (4.72%DM) and low (2.32%DM) tannin grain sorghum based diets, respectively. Each GSV were supplemented with 0% (control, 10%DM-SCM and 10%DM-MOLM (SP_1, SP_2 and SP_3 respectively). Birds were randomly allocated to the 6 dietary treatment groups of 45 birds each. Each treatment group was further subdivided in 3 replicates of 15 birds each. Each treatment replicate was randomly placed in a single pen in poultry house for eight weeks.

At the end of the eight (8) weeks of feeding trial, two birds were randomly selected from each treatment replicate; six birds per treatment making a total of 36 birds for the six dietary treatments and were randomly placed in battery cages. The sampled birds were starved for 6 hours before taking blood samples. The birds were bled early in the morning between 6.30 and 7.30 using a sterilized disposable syringe and a needle. Birds were punctured at the wing vein to aspirate blood. A total of 6ml of blood was sampled from each bird of which 2mls were placed into bijon bottle treated with ethylene diamine tetra acetic acid (EDTA) for hematological assay. The remainder (4ml) of the blood sample was allowed to coagulate to produce sera for blood chemistry measurements according to the methods of Okeudo *et al.* (2003). Blood samples were analyzed within 3 hours after their collection for total erythrocyte, haematocrit (PCV), haemaglobin (Hb) and differential leucocytes count according to the methods described by Dein, (1984).

Erythrocyte (RBC) count was done in a haemo-cytometer chamber with Natt and Herdrics diluents' to obtain a 1:200 blood dilution. Packed cell volume (PCV) was measured as micro haematocrit with 75 x 16mm capillary tubes filled with blood and centrifuged at 3000 rpm for 5 minutes. Micro-haemotocrit reader was used to estimate the percent of PCV in blood. The differential count of leucocytes was made from blood stained with Wrights dye and each type of cell countered using a laboratory counter under 100 microscopic magnification object. Percent haemaglobin (HB) was measured using sahli method. Percent of mean cell haemaglobin concentration (MCHC) was measured as proportion of HB and PCV. Coagulated blood was subjected to the standard method of serum separation of the harvested total serum protein (TSP) using Biuret method end point and expressed as g/dl. Albumin concentration was determined by the Bromocresol Green (BCG) Dye method described by Peters *et al.* (1982) and expressed as g/dl. Globulin (GB) concentration was computed as the difference between total protein and albumin concentrations. Glucose concentration in serum was determined by the Glucose oxidase/peroxidase method and expressed as mg/dl. Cholesterol in serum samples were determined by chod-pap method and expressed as mg/dl.

Data analysis

All data collected on hematological parameters and serum biochemical indices for GVS and SPS was analyzed in accordance with the 2x3 factorial designs using the General Linear Models procedure of SAS software version 9.1 for windows (2007). Values were considered significant at ($P \leq 0.05$). The Least Square Difference was used to compare means of each variable. The analytical model for studied haematological parameters and serum biochemical indices was as follows: $Y_{ijkl} = \mu + V_i + S_j + R_l + (VS)_{ij} + e_{ijk}$

Where:

Y_{ijk} = Observation of k^{th} bird assigned to i^{th} GSV subjected to $_{jth}$ SPS;

μ = overall mean common to all observations

V_i = effect of GSV based diet (HTS or LTS)

S_j = the effect of SPS based diet (CTL, SCM or MOL);

R_l =the effect of treatment replicates (Replication, 1,2 or 3

$(VS)_{ik}$= the interaction between GSV and SPS;

e_{ijk} = random error peculiar to each observation

Results

Effects of grain sorghum

The results of haematological parameters amongst GSV dietary groups are shown in (Table 2). Hb, MCHC and neurophil ratio values were not affected by type of GSV. The RBC and WBC concentration and lymphocyte and oesinophil ratio values were higher in HTS and lower in LTS dietary group. However, the monocyte ratio was higher in LTS and lower in HTS dietary group. The results of metabolite indices amongst GSV dietary groups are shown (Table 2). The serum glucose, albumen, globulin, and cholesterol concentration values were not affected by GSV. Though, the cholesterol concentration was slightly lower in HTS and higher in LTS dietary group but the difference was not significant.

Effects of supplementation

The results of hematological and serum metabolite parameters amongst SPS dietary groups are shown in (Table 3). The addition of SCM and MOLM in GSV diets had no significant effect on RBC, Hb, MCHC, WBC concentration and PCV and basophil and neutrophil ratios, serum albumen, globulin and cholesterol concentration values. Addition of SCM and MOLM decreased lymphocyte ratio values compared to CTL. However, addition of SCM in GSV diets decreased more lymphocyte ratio values compared with MOLM addition. Moreover, the supplementation of SCM did not change oesinophils ratios but MOLM decreased oesinophils ratios. The supplementation of SCM increased TSP whereas MOLM supplementation did not change TSP value.

The interaction between type of GSV and type SPS on lymphocyte, monocyte and oesinophil ratios are shown in (Table 4). The lymphocyte ratio was higher in HTS and lower in LTS dietary group. The supplementation of SCM and MOLM in HTS based diet decreased lymphocyte ratio. The supplementation of SCM in LTS based diet decreased but supplementation of MOLM did not change lymphocyte ratio value. Furthermore, oesinophil ratio value was higher in HTS and lower in LTS dietary group. The supplementation of SCM and MOLM in HTS based diet decreased oesinophil ratio value. On the contrary, the supplementation of sugar cane molasses and moringa leaf meal in LTS based diet did not change the value of oesinophil ratio. Moreover, the monocyte ratio value was higher in HTS and lower in LTS dietary group. But, supplementation of sugar cane molasses and moringa leaf meal in HTS based diet significantly increased the monocyte ratio value compared to CTL_1. Similarly, supplementation of sugar cane molasses and moringa leaf mea in LTS based diet significantly increased monocyte ratio value compared to CTL_2. Nevertheless, the supplementation of sugar cane molasses in LTS based diet increased significantly increased the monocyte ratio value compared with supplementation.

Discussion

The PCV, Hb and MCHC values among GSV dietary groups in the present study were within the range values reported by other authors (Islam *et al.* 2004 and Medugu *et. al.* 2010). The similarity observed amongst HTS and LTS dietary groups on PCV, Hb and MCHC values agree with those workers (Kwari *et al.* 2011; Kwari *et al.* 2012). However, these results were in conflict with Medugu *et al.* (2010) who observed lower PCV and WBC in HTS and higher Hb and MCHC in HTS. The differences in the results were probably due to the differences in the levels of dietary tannins in the two studies. High tannin levels have been reported to adversely affect PCV, Hb and MCHC (Medugu *et al.* 2010 and Etuk *et al.* 2012). These findings suggest that probably the level of dietary tannin in the present study was not high enough to exert negative effects on these attributes. The results suggest up to 2.6%DM as leucocynidin equivalent has no detrimental effect on hematological parameters.

The RBC and WBC values observed among GSV dietary groups in the present study compare well with those other researchers (Medugu *et al.* 2010; Kwari *et al.* 2014). Higher RBC and WBC values noted in HTS compared to LTS dietary group were expected. High dietary tannin levels have been reported to negatively affect RBC and WBC whereas low dietary tannins have no detrimental effects on these hematological parameters (Medugu *et. al.* 2010; Etuk *et al.* 2012). These results could be attributed to the nutritional stress in HTS due to the negative effect of tannins. Nutritional stress results into increase in various types of WBC such as neutrophil, band cells (slightly immature neutrophil), T-type lymphocytes (T cells), B-type lymphocytes (B cells), monocyte, oesinophils and basophiles to appear as response to nutritional inadequacy. According to Payne *et al.* (1990) shortage of dietary proteins and amino acids alters immune response. Moreover, the nutritional stress may be resulted into an increase of RBC production to compensate for reduced oxygen carrying capacity associated with inadequate protein such as globulins due negative effects of tannins. These findings suggest the use up to 2.6%DM of HTS based diets in growing chicks lead to nutritional stress, reduce oxygen carrying capacity and ultimately could negatively affect health of the birds. Thus, use of HTS based diet in growing chicks should be accompanied with protein and energy supplementation.

The differential leukocyte count values observed among GSV dietary groups were within the range reported by other researchers (Medugu *et al.* 2010; Islam *et al.* 2004). However, the higher lymphocyte and oesinophil ratios observed in HTS than in LTS dietary group was an indication of the response to nutritional stress in HTS dietary group. These findings could be attributed to inadequacy of nutrients such as energy and amino acids in HTS due to the negative effects of tannins (Gross and Siege, 1983; Kwari *et al.* 2010). The lymphocyte and oesinophil increase with increase in stressors to birds (McLane and Curtis, 1989). The increase in lymphocyte and oesinophil ratio has been a reliable indicator of stress in chickens (Cross and Siegel, 1983; Macframe and Curtis, 1987). These findings suggest that the use of HTS in the diets of growing chicks could suppress immune functions of growing chicks. Moreover, the decrease of monocyte ratios in HTS compared to LT-GS dietary group could be attributed to inadequacy of nutrients responsible for monocyte formation in HTS due to the negative effect of tannins. The inadequacy of nutrients may suppress the proliferation of monocyte in HTS dietary group. These results agree with those of Medugu *et al.* (2010) and Kwari *et al.* (2011).Thus, utilization of HTS based chick diets could lead to disruption of humeral and cell mediated immunity in growing chicks. Therefore, use of HTS based should be accompanied with supplementation of nutrients such as amino acids and energy.

Serum glucose is commonly referred to as blood sugar or it refers concentration of sugar or glucose in the blood stream. The similarity of mean serum glucose values observed between HTS and LTS dietary groups is in agreement with those obtained in previous studies (Medugu *et al.* 2010; Kwari *et al.* 2011). These results might be due to the characteristics of birds that appear to maintain high and relatively constant blood glucose values, even when in low feed intake (Liukkonen-Anttila, 2001). These findings suggest that tannin level has no influence on blood glucose level.

The moderate decreases in serum cholesterol concentration observed in HTS compared to LTS dietary group suggest that tannins have hypocholesterolemic properties. These findings may be explained by the metabolic and physiological actions of tannins. Tannins promote the excretion of fecal sterols, thereby leading to a decrease in dietary cholesterol as well as plasma and hepatic cholesterol (Park *et al.* 2002). Tannins also increase fiber content which block the intestinal cholesterol absorption and decrease serum cholesterol (Lansky *et al.* 1993). The findings suggests the use HTS based chick diets could reduce cholesterol concentration in growing chickens and demonstrate that the use of tannin products in poultry could be beneficial for production of low cholesterol poultry products and reduce risks of cardio-vascular diseases in humans. These results are in agreement with other researchers elsewhere (Kwari *et al.* 2011; Park *et al.* 2012).

The comparable values of serum total protein, albumen and globulin concentration observed in HTS and LTS dietary groups indicate that there were no negative effects of tannin on protein component in the present study. The reason for these findings was not clear but could probably they could be attributed to the higher feed intake observed in HTS dietary group which probably led to higher feed intake and hence offset the negative effect of tannins on those attributes. These results are congruent with those of Kumar *et al.* (2007). These findings suggest that up to 2.6%DM tannin concentration may not be detrimental to total blood proteins, albumen and globulins.

The SPS dietary groups did not differ with respect to PCV, Hb, and MCHC and WBC values. This was an indication of the adequacy of nutrients responsible for formation of blood variables amongst CTL dietary groups. These findings suggest that supplementation of molasses and MOLM to GSV based diet could not exert the detrimental effects on hematological variables. The range in lymphocyte and oesinophil ratio values observed amongst SPS dietary groups was within values reported from other research work (Islam *et. al.,* 2004; Medugu *et al.* 2010). However, the decrease of lymphocyte and oesinophil ratio observed in SCM and MOL dietary groups was an indication of nutritional stress in birds fed GSV diets (McLane and Curtis, 1989). Therefore, supplementation of molasses and MOLM to GSV based diet compensates deficient nutrients; hence improve the nutritional status of growing chicks. However, the interaction between GSV and SPS observed in the present study on lymphocyte and oesinophil was indicating differences of two GSV on nutrient content due to their differences in tannin content. The higher lymphocyte and oesinophil in CTL_1 than CTL2 dietary group indicating nutritional stress in HTS due to negative effect of tannins and nutritional adequacy in LTS dietary group. The decrease of lymphocyte and oesinophils values in SCM_1 and MOL_1 dietary group were within the values reported by Islam *et al.* (2004). Thus, may be supplementation of SCM and MOLM to HTS based diets compensated the deficient nutrients, alleviated nutritional stress and improved immunity. However, the insignificant change of lymphocyte and oesinophils values in MOL_2 was showing nutritional adequacy in LTS. But the significant decrease of lymphocyte in SCM_2 dietary group may be could to be attributed to inadequate nutrients due negative effects of SCM. These findings show that supplementation of MOLM and SCM to HTS diets could alleviate the negative effects of tannins and improve nutritional status whereas supplementation of SCM to LTS could suppress nutritional status.

Monocytes are a type of leukocytes or white blood cell which plays a role in immune system function. The monocyte move to the body tissues and become macrophages and directly perform phagocytosis. The range of monocyte ratio values observed among SPS dietary groups were wider compared to those values reported by other researchers (Islam *et al.* 2004; Medugu *et al.* 2010). The insignificant change of monocyte ratio in MOL dietary group suggests the absence of foreign bodies and toxic materials in MOL which might be detrimental to health of birds. However, the significant interaction observed between SPS and GSV on lymphocyte ratio was an indication of the differences in nutrients between two GSV and is associated with their difference in tannin levels. Supplementation of MOL and SCM to HTS diets increased monocyte ratios indicating the deficiency of nutrients in the GSV due to negative effects of tannins. These findings could be attributed to the compensation of deficient nutrients in HTS diets by MOL and SCM. Thus, these findings suggest that supplementation MOL and SCM to HTS improve humeral and cell mediated immunity for growing chickens. However, supplementation of SCM to LTS increased monocyte ratio indicating of deficient nutrients in SCM dietary group due to the negative effect of SCM in utilization nutrients. Thus, utilization of SCM as an additive to LTS based diets is not beneficial because could disrupt the immune response of birds.

The serum glucose levels observed amongst SPS dietary groups in the present study were lower compared with those from other research reports (Imik *et al.* 2006; Etuk *et al.* 2012; Ahmed *et al.* 2013). The reason for the contrasting results could be associated with differences in age, breeds, type of energy sources and ingredients used in different studies. The insignificant difference of serum glucose concentration observed among SPS dietary groups could be associated with hypoglycemic nature of GSV, molasses and MOL (Kim and Park, 2012). The non-significant increase of serum glucose in MOL dietary groups demonstrates that MOL has hypoglycemic properties. Moringa leaves contain pyto-chemicals or specific inhibitor of intestinal sucrose that reduce blood sugar (Adisakwattana and Chanathong, 2011). In addition, the aqueous extract of MOL normalize elevated hepatic pyruvate carboxylase enzyme and regenerate damaged hepatocytes and pancreatic β cells via its antioxidant properties to posses the hypoglycemic effect (Amira *et al.* 2014). Thus, these findings suggest that the supplementation of MOL could reduce serum glucose and inhibit over fattening to improve health of growing chicks. Further, the utilization of MOL may result into desirable poultry products with less sugar and fats particularly to diabetic patients. In addition, slight increase of serum glucose observed SCM dietary group may suggest the glycemic index (GI) lowering properties of molasses. The molasses extract may contain one or more of the following substances: lipids, phospholipids, protein, flavonoids such as anthocyanins, catechins, chalcones, flavonols and flavones, polyphenols, antioxidants, phytosterols such as 1-octacosanol, campesterol, stigmasterol, β-sitosterol, oligosaccharides such as raffmose, 1-kestose, theanderose, 6- kestose, panose, neo-kestose and nystose, and organic acids such as c-aconitic acid, citric acid, phosphoric acid, gluconic acid, malic acid, t-aconitic acid, succinic acid and lactic acid, aliphatic alcohols, vitamins, minerals, carbohydrates, gums and neutral and polar lipids. Thus, these findings suggest utilization of sugar cane molasses could inhibit over fattening in growing chicks and potential low sugar poultry products particularly for diabetic patients.

The moderate decrease of serum cholesterol concentration with supplementation of sugar cane molasses and moringa leaf meal to GSV based diet were suggesting that molasses and MOLM have hypocholesterolemic properties (Lansky *et al.* 1993; Olugbemi *et al.* 2010). Thus, the use of molasses and MOL could produce low cholesterol poultry products. These results agree with other research reports elsewhere (Njidda *et al.* 2006; Ghazalah and Ali, 2008; Olugbemi *et al.* 2010). The hypocholesterolemic properties in molasses can be explained by series of n-alkenes (C_{23}-C_{33} and ethyl and methyl esters of fatty acids mainly (oleate and palmamitate) of phytosterols (stigma- sterol, campestrols, and β-sito-sterols), free fatty acids, triglycerides and keto-steroid derivatives contained in molasses. They may inhibit the intestinal cholesterol absorption and thereby decrease plasma total LDL cholesterol levels (Ghasi *et al.* 2000; Goldstein *et al.* 2000). Moreover, the phytosterols present in molasses that affect cholesterol metabolism and interfere with steroid synthesis (Goldstein *et al.* 2000). Furthermore, the hypocholesterolemic properties of MOL may be associated with high fibrous material that block the intestinal cholesterol absorption and ultimately decrease the serum cholesterol or the presence bioactive such as phytochemicals, the phytosterols such as β-sito-sterols, campe-sterols, stigma-sterols and avenasterol present in MOL that decrease the plasma concentration of low density lipoproteins (LDL) cholesterol (Ghasi *et al.* 2000; Anwar *et al.* 2007). The sterols inhibit body cholesterol production in the liver. But, the phytosterols affect aspects of cholesterol metabolism and interfere with steroid synthesis (Goldstein *et al.* 2000). Thus, these findings suggest utilization of SCM and MOL as additive to GSV based diets for growing chicks reduce blood cholesterol levels and suggesting a potential to produce low cholesterol poultry products and that could reduce health risks associated with high blood cholesterol such as cardio-vascular diseases.

The total serum protein measures the amount of albumin and globulins in blood. The albumin prevents leakage of blood vessels whereas globulin maintains the immune system. The total serum protein levels observed amongst SPS dietary groups in the present study compare well with results reported from in other research work (Medugu *et al.* 2010; Ahmed *et al.* 2013). The significant increase of total serum protein levels in MOL diets compared to CTL dietary group was an indication of low level of quality protein in GSV. The total serum protein reflects the protein quality fed (Eggum, 1970). These findings may be indication of substantial amount of quality proteins in MOL. Thus, addition of MOL to GSV increased the plasma protein levels which compose of mainly three proteins, globulin, albumen and fibrinogens. The plasma proteins play an important role in regulation of the body's osmotic pressure. They also help to maintain fluid and electrolyte balance in blood, thus keeping the body's functions working properly. Degeneration of plasma proteins can cause health problems such as distended blood vessels and disruption of immune system. These findings suggest that addition of MOL to GSV based diets could improve the amount of quality of protein and eventually improve health and immunity in growing chicks. These results concurs with those of other workers (Onu and Aniebo, 2011;Tesfaye and Zeit, 2013).However, the non-significant change of total serum proteins observed in SCM compared to CTL dietary group was an indication of presence of low quality proteins in molasses. Thus, addition of molasses has no influence on total serum proteins. Moreover, the non-significant difference among SPS dietary groups on albumen and globulins indicate that addition of molasses and MOLM to GSV based diets had no influence on these attributes.

Conclusions

- It can be concluded that use of HTS based diets can disrupt immune system and lead to nutritional stress in growing chicks due to the negative effect of tannins. Thus, its utilization should be accompanied with supplementation of energy, proteins and amino acids to alleviate the negative effects of tannins and improve immune response and nutritional status of growing chicks.
- The supplementation of MOLM and SCM to HTS diets can alleviate the nutritional stress and humeral and cell mediated immunity in growing chicks. Thus, up to 10%-DM supplementation of MOLM and SCM in HTS based chick diets are suggested.
- The supplementation of SCM in LTS based diets can disrupt immune response in growing chicks. Thus, its utilization for improvement of feeding value of LTS for growing chicks is not beneficial.

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Table 1: Composition of experimental diets

Ingredient	D_1	D_2	D_3	D_4	D_5	D_6
Moringa leaf meal	-	-	-	-	11.3	11.3
Molasses	-	-	14.4	14.4	-	-
Sorghum meal (High Tannin)	62.5	-	61.4	-	61.4	-
Sorghum meal (Low Tannin)	-	62.3	-	61.4	-	61.4
Fish meal	16.0	16.0	16.0	16.0	16.0	16.0
Sunflower seed meal	16.7	16.9	16.9	16.9	16.9	16.9
Limestone	2.00	2.00	2.00	2.00	2.20	2.20
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Bone meal	2.00	2.00	2.00	2.00	2.00	2.00
Salt	0.40	0.40	0.40	0.40	0.40	0.40
Chemical Composition (%)						
Dry matter	89.6	89.5	87.1	88.3	89.0	89.4
Crude protein	23.6	24.5	21.6	22.5	26.1	26.4
Crude fiber	9.60	9.27	7.41	6.50	10.6	8.03
Ether extract	7.66	8.46	5.97	6.76	7.38	6.87
NFE (Starch +sugar)	47.1	47.2	51.2	52.2	39.7	39.0
Lysine	1.00	1.11	1.03	1.38	1.40	1.54
Methionine &cystine	0.86	0.67	0.70	0.77	0.75	0.82
Tryptophan	0.23	0.28	0.29	0.31	0.26	0.28
Tannin	2.60	1.30	2.15	1.05	2.55	1.36
ME (MJ/kgDM)	10.8	11.1	12.05	12.4	11.7	12.0

D_1-HTS based diet (CTL_1); D_2- LTS based diet (CTL_2); D_3- HTS+ molasses (SCM_1); D_4- LTS+ molasses (SCM_2); D_5- HTS+MOLM (MOL_1); D_6- LTS+MOLM (MOL_2).

Table 2: Blood and serum metabolite parameters of growing chicks fed high and low tannin grain sorghum varieties (0-8 weeks of age)

	Grain sorghum variety based diets			
	HTS	LTS	SED	Prob
PCV (%)	29.2	28.2	2.31	0.381
Hb (%)	8.04	8.16	0.988	0.816
MCHC (%)	27.7	28.9	3.49	0.472
RBC ($x10^6/\mu$)	2.71 [a]	1.65 [b]	0.61	0.030
WBC ($x10^6/\mu$)	8.42 [a]	5.68 [b]	1.71	.0394
Lymphocyte (%)	65.3 [a]	51.2 [b]	6.073	0.0099
Neutrophils (%)	20.3 [a]	23.3 [a]	6.807	0.478
Oesinophils (%)	6.00 [a]	1.50 [b]	6.807	1.51
Monocyte (%)	6.83 [b]	19.3 [a]	3.59	0.0018
Total serum protein (g/dl^{-1})	2.38	3.26	1.92	0.356
Globulin (g/dl^{-1})	0.184	1.093	1.68	0.277
Albumen (g/dl^{-1})	2.20	2.17	0.441	0.886
Glucose (mg/dl^{-1})	93.2	80.9	19.9	0.230
Cholesterol (mg/dl^{-1})	92.4	142	88.5	0.262

[ab] *Means in the same row with different superscripts are different ($P \leq 0.05$)*; SED = Standard error of difference; HTS.55%DM-high tannin grain sorghum based diet; LTS- 55%DM-low tannin grain sorghum based diet.

Table 3: Blood and serum metabolite parameters of growing chicks fed MOLM and molasses supplemented grain sorghum based diets (0-8 weeks of age)

Parameter	Supplementary diet				
	CTL	SCM	MOL	SED	Prob
Packed Cell Volume (%)	28.5	28.0	29.7	2.307	0.466
Haemoglobin (%)	8.33	8.27	7.70	0.988	0.499
MCHC (%)	29.2	29.6	26.1	0.223	3.490
RBC ($x10^6/\mu$)	2.19	2.208	2.14	0.61	0.986
WBC ($x10^3/\mu$)	7.17.5	6.88	7.76	1.71	0.60
Lymphocyte (%)	72.0 [a]	44.8 [c]	58.00 [b]	6.073	0.0041
Neutrophils (%)	22.0	26.0	17.5	6.807	0.297
Oesinophils (%)	5.25 [a]	3.50 [a]	2.50 [b]	1.51	0.117
Monocytes (%)	7.00 [b]	25.8 [a]	6.50 [b]	3.59	0.0010
Total serum protein (g/dl^{-1})	1.87 [b]	4.79 [a]	1.809 [b]	1.92	0.0359
Albumen (g/dl^{-1})	1.87	2.56	2.98	0.441	0.0831
Globulin (g/dl^{-1})	0.0149	0.0173	2.24	1.68	0.0506
Glucose (mg/dl^{-1})	78.7	97.1	85.3	19.9	0.3101
Cholesterol (mg/dl^{-1})	168	88.5	95.2	88.5	0.508

[ab] *Means in the same row with different superscripts are different ($P \leq 0.05$)*; SED = Standard error of difference; CTL.55%DM-grain sorghum+'0' supplementation; SCM.55%DM-grain sorghum plus 10%DM' molasses supplementation; MOL.55%DM-grain sorghum plus10%DM-MOLM supplementation.

Table 4: Interaction of tannin levels in grain sorghum variety and type of supplementary strategy on lymphocytes, oesinophils and monocyte ratio of growing chicks at 8 weeks of age.

Parameter	HTS			LTS				
	CTL_1	SCM_1	MOL_1	CTL_2	SCM_2	MOL_2	SED	Prob
Lymphocytes	77.0^{a}	60.0^{b}	50.0^{b}	67.0^{b}	29.5^{c}	57.0^{b}	6.073	0.0489
Oesinophils	9.50^{a}	5.50^{b}	3.00^{bc}	1.00^{bc}	1.50^{bc}	2.00^{bc}	1.51	0.0437
Monocytes	2.00^{bc}	11.5^{b}	7.00^{bc}	12.0^{b}	40.0^{a}	6.00^{bc}	3.59	0.0489

^{abc} *Means in the same row with different superscripts are different ($P \leq 0.05$)*; SED = Standard error of difference; CTL_1-55%DM- high tannin grain sorghum+'0' supplementation;CTL_2 -55%DM- low tannin grain sorghum+'0' supplementation; SCM_1-55%DM- high tannin grain sorghum plus 10%DM' molasses supplementation; SCM_2-55%DM- low tannin grain sorghum plus 10%DM' molasses supplementation; MOL_1-55%DM- high tannin grain sorghum plus10%DM-MOLM supplementation; MOL_2-55%DM- low tannin grain sorghum plus10%DM-MOLM supplementation

CHAPTER SIX

6.0 PAPER IV

Moringa oleifera leaf meal and molasses as additives in grain sorghum based diets for layer chickens.

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Moringa oleifera leaf meal and molasses as additives in grain sorghum based diets for layer chickens

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Abstract

The effects of supplementing *Moringa oleifera* leaf meal and molasses to grain sorghum based diets for commercial layer chickens on egg production was studied in a 2x3 factorial design for ten weeks. The study used high (HT) and low (LT) tannin varieties of sorghum and three supplement strategies of: none, molasses (Mo) or Moringa leaf meal (ML).

Egg weight was not influenced by tannin level. Daily feed intake was higher, feed conversion poorer; egg laying percentage and egg mass production were lower in HT than in LT diets. Supplementation with molasses depressed, while supplementation with Moringa leaf meal improved, egg production. Final sugar cane molasses is not recommended as additive to sorghum based diets as it may depress egg production and impair feed conversion. *Moringa oleifera* leaf meal appears to improve egg production on sorghum based diets and may reduce the expense of using costly synthetic amino acids. Its utilization as an additive in sorghum-based diets is highly recommended.

Key words: *agro forestry, egg production, tannin, Tanzania*

Introduction

Sorghum *(Sorghum bicolor*) is well adapted to arid and semi-arid regions and could be used as an alternative to maize due to its high energy content and its chemical composition is similar to maize, except for the deficiency in lysine, methionine, and threonine (USDA 2009 and Mutayoba et al 2011). Sorghum ranks the fifth as the most produced crop worldwide (NRC 1996). The use of sorghum in poultry diets is limited probably due to the presence of tannin in some of the varieties (Dykes and Rooney 2006; Medugu et al 2010). The Proanthocyanidins (condensed tannins) consisting of oligomers and polymers of flavan-3-ol units are the most widely distributed type of tannins in plants (Waterman and Mole 1994). In rare cases, Proanthocyanidins comprising 3-deoxy subunits also exist particularly in sorghum and maize. Studies have shown that the presence of these compounds at more than 1% in the diet could depress egg production performance in layers (Potter et al 1967; Armstrong et al 1973). In addition, excessive tannin consumption can lead to a reduction in feed intake, egg weight, laying rate and poor feed conversion ratio (Sell et al 1983; Kyarisiima et al 2004). Tannin is thought to impair digestion and absorption of nutrients (Lasheras et al 1980a, b), alter nutrient partitioning and tissue composition (Cherian et al 2002; Du et al 2002), and cause cell degeneration in the liver and kidneys (Harvey and McAllan 1992).

Various studies to improve nutritional quality and utilization of high-tannin grain sorghum using physical and chemical treatment (Mitaru et al 1983 and Elkin et al 1990) and dietary nutrient enrichment (Kumarl et al 2005) have been carried out. Furthermore, attempts to increase dietary protein and energy levels using common ingredients in high tannin grain sorghum based diets have also been done (Sell et al 1983; Pour-Reza and Edriss 1997). Both nutritional strategies were found to improve partially performance in laying birds. It is anticipated that the use of *Moringa oleifera* leaf meal and molasses that are readily available and cheap in tropical countries could alleviate the negative effects of tannins on egg production performance since they contain essential nutrients with negligible anti-nutritional factors (Alikwe and Omotosho 2003; Aye and Adegun 2003). However, there is limited information on the effect of using Moringa leaf meal and molasses in high tannin grain sorghum based diets on egg production. Thus, the objective of this study was to find out if supplementation *of Moringa oleifera* leaf meal or molasses to high tannin grain sorghum based diets could alleviate the undesirable effects of tannins and improve laying performance.

Materials and Methods

Feed ingredients and experimental diets

The study was conducted at Sokoine University of Agriculture (SUA) at the Poultry Unit in the Department of Animal Science and Production (DASP) in Morogoro, Tanzania. *Moringa oleifera* leaves were harvested within SUA compound, air dried under a shed to maintain their greenish colour, ground with hammer mill to produce Moringa leaf mal (ML). Grain sorghum varieties were collected from the local markets, ground with hammer mill to pass through a 2mm sieve. Molasses was obtained from Mtibwa sugar processing company. The methods of Association of Official Analytical Chemists (AOAC 1990) were used to determine proximate composition and minerals of the feed ingredients and experimental diets. Metabolizable Energy (ME/ kcal/kg) content of feed ingredients and experimental diets were estimated by prediction equations expressed by NRC (1994). MOLM, ME/Kgcal/kg was estimated by prediction equation established by Carpenter and Clegg (1956). Condensed tannins in GSV and MOLM were determined using butanol/HCL method as described by Nitao et al (2001). All ME_n values were converted into MJ/kg DM. All diets were formulated to meet or exceed the nutrient requirements recommended by NRC (1994) (Table 1) for layer chickens. The inclusion of the sorghum was fixed at 55% of the DM.

Experimental design and dietary treatments

Six dietary treatments (Table 1) were arranged in a 2 x 3 factorial design.
The factors were:
Tannin: Low (LT) or high (HT)
Supplement (SPS): None (CTL), molasses (Mo) or Moringa leaf meal (ML)

Experimental procedure

Two hundred and seventy (270) commercial Bovan brown hybrid layer chickens at 20th week of age were randomly allocated to the dietary treatments. Each dietary treatment had 45 birds subdivided into three replicates of 15 birds each. The birds were housed in deep litter pens. The birds were provided with the experimental diets from the 20^{th} to 30^{th} week of age. Prior to the commencement of the experiment, each bird was weighed individually to obtain the initial body weight and at the end of experimental period to get final weight. Birds were group fed in each replicate. The experimental diets supplied to the birds were weighed daily and were given to the birds every morning at 8.00A.M. The amount of feed offered daily was approximately 20 percent above the expected intake. The remaining feed (residue) was also weighed daily before supplying fresh feed. Feed intake per bird was calculated by subtracting the leftovers from feed and was divided by the number of birds. Eggs were collected and recorded on a daily basis. Eggs were weighed daily using sensitive digital weighing scale. Daily laying percentage was calculated as the number of eggs produced over a period of time by dividing the number birds x 100. Average egg weight was calculated as total egg weight by dividing the number of recorded eggs. Daily egg mass was calculated as average egg weight multiplying by daily laying percentage. The daily feed intake (g) over daily egg mass (g) was used to calculate the feed conversion ratio (FCR).

Data analysis

All data collected on egg production performance parameters for GVS and SPS were analyzed in accordance with the 2x3 factorial designs of two GSV in combination with three SPS using General Linear Model procedure of SAS software version 9.1 for windows (2007). Values were considered significant at ($P \leq 0.05$). The Least Square Difference was used to compare means of each variable. The analytical model for studied parameters were as follows: $Y_{ijk} = \mu + V_i + S_j + (VS)_{ij} + e_{ijk}$

Where:

Y_{ijk} = Observation of k^{th} bird assigned to i^{th} GSV subjected to $_{jth}$ SPS;

μ = overall mean to all observations;

V_i= effect of tannin (HTS or LTS);

S_j= the effect of supplement: CTL (no supplement), Mo (molasses) or ML (Moringa leaf meal)

$(VS)_{ik}$= the interaction between GSV and SPS;

e_{ijk} = random error

Table 1: Composition of experimental diets

Ingredient	HT	LT	HT-Mo	LT-Mo	HT-ML	LT-ML
Moringa leaf meal	-	-	-	-	11.3	11.3
Molasses	-	-	14.4	14.4	-	-
Sorghum meal (high tannin)	62.5	-	62.5	-	62.5	-
Sorghum meal (low tannin)	-	62.3	-	62.3	-	62.3
Fish meal	10.0	10.0	10.0	10.0	10.0	10.0
Sunflower seed meal	18.0	18.0	18.0	18.0	18.0	18.0
Limestone	6.50	6.80	6.50	6.80	6.50	6.80
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Bone meal	2.25	2.25	2.25	2.25	2.25	2.25
Salt	0.50	0.50	0.50	0.50	0.50	0.50
Chemical composition (%DM)						
Dry matter	88.9	90.6	87.8	87.0	89.5	88.6
Crude protein	17.0	16.7	17.9	19.3	20.5	19.8
Crude fiber	9.60	9.27	7.41	6.50	10.6	8.03
Ether extract	7.52	9.51	5.62	6.34	9.08	9.85
NFE (Starch +sugar)	33.7	34.9	39.1	39.8	39.0	39.3
Lysine	0.885	0.868	1.091	1.101	3.102	3.12
Methionine &cystine	0.5087	0.499	0.655	0.6609	1.054	1.065
Tryptophan	0.186	0.183	0.213	0.215	0.263	0.265
Tannin	2.60	1.30	2.15	1.05	2.55	1.36
ME (MJ/kgDM)	9.42	8.77	8..606	9.23	9.27	9.105

HT-.55%DM-High tannin grain sorghum based diet; LTS- 55%DM-Low tannin grain sorghum based diet; HT-Mo- 55%DM-High tannin grain sorghum+10%DM-molasses; LT-Mo- 55%DM-Low tannin grain+10%DM-molasses; HT-ML- 55%DM-High tannin grain sorghum+10%DM-Moringa leaf meal; LT-ML- 55%DM-Low tannin grain sorghum+10%DM-Moringa leaf meal.

Results

Effect of tannin

The egg weight (EWT) values did not differ between HT and LT diets (Table 2). The laying percentage (DLP) and egg mass production (EMP) were lower by 32.2 and 27.9% respectively in HT compared to LT dietary groups. The daily feed intake (DFI)) was higher by 7.2% and the feed conversion poorer by 59% in HT compared to LT dietary groups, respectively.

Effect of supplement

The DLP value decreased with molasses by 21.1% but increased with Moringa by 8.7% (Table 3). Similarly, the EMP value was decreased by 19.9% in molasses supplemented group but increased by 7.24% in MOL supplemented group. The DFI increased by 2.63% with molasses but did not change with Moringa, compared to the control. The FCR value did not differ among supplements. There was an interaction between tannin diets and supplements (Table 4). Molasses had a negative effect on feed conversion when it was added to the low tannin diets but had no effect in the high tannin diets.

Interaction of tannin and supplement

The interaction of tannin level and supplementary strategies on FCR are shown in Table 4. Supplementary of SCM and MOL to HTS decreased FCR by 27.6% and

29% respectively. However, supplementary of SCM to LTS increased FCR by 156% whereas supplementation of MOL did not change FCR.

Table 2: Egg production, feed intake and feed conversion ratio of bovan hybrid layers fed high and low tannin grain sorghum varieties (20-30 weeks of age)

	Grain sorghum variety based diets			
	HT	LT	SEM	Prob
Daily laying percentage, %	31.2^{b}	46.1^{a}	5.45	0.0002
Average egg weight, g	53.1	52.8	1.29	0.58
Daily egg mass production, g/bird	17.3^{b}	24.0^{a}	2.82	0.002
Daily feed intake, g/bird	119.0^{a}	111.0^{b}	2.92	0.0003
Feed conversion, g feed/g egg mass	11.0^{a}	6.93^{b}	2.83	.0047

^{ab}*Means in the same row without common superscripts are different at $P \leq 0.05$; SEM = Standard error of difference*

Table 3: Mean values for egg production feed intake and feed conversion of commercial bovan hybrid layers fed Moringa leaf meal and molasses supplemented grain sorghum based diets (20-30 weeks of age)

	CTL	SCM	MOL	SEM	Prob
Laying percentage, %	40.3^{b}	31.8^{c}	43.8^{a}	5.45	0.0094
Egg weight, g	53.2	52.7	53.0	1.29	0.80
Egg mass, g/bird/day	22.1^{b}	17.7^{c}	23.7^{a}	2.82	0.011
Feed intake, g/day	114^{b}	117^{a}	114^{b}	2.92	0.135
Feed conversion, g feed/g egg mass	9.53	11.1	7.38	2.83	0.123

^{ab}*Means in the same row without common superscripts are different at $P \leq 0.05$*

Table 4: Interaction of tannin levels in grain sorghum variety and type of supplementary strategy on feed conversion

	HTS			LTS				
	CTL	Mo	ML	CTL	Mo	ML	SEM	Prob
Feed conversion	14.5^{a}	10.5^{a}	10.3^{a}	4.58^{b}	11.72^{a}	4.50^{b}	2.83	0.0202

^{abc}*Means in the same row with different superscripts are different (P*

Discussion

The lower DLP in HT compared to LT dietary groups concur with other research reports (Faquinello et al 2004; Kyarisiima 2004). These findings were attributed to the negative effects of tannins on utilization and digestibility of nutrients such as protein and amino acids responsible for egg formation (Hassan et al 2003; Ravindran et al 2006). These results support previous studies that tannin levels between 1.5% and 3% may depress egg production (Ali and Mahmood 2003). According to Potter and Fuller (1968), HT based diets have a depressive effect on DLP and its use should be accompanied with supplementation of proteins and amino acids particularly methionine and choline which donate a methyl group that combines with tannins and render them ineffective.

The influence of tannin level on feed intake in layers is inconsistent in the literature. The higher value in HT compared to LT dietary group agrees with other research reports (Nyakoti and Atkinson 1995; Nyakoti et al 1997). These findings are associated with the compensatory mechanism of birds to increase intake due to the lower energy concentration of the high tannin diet (Cherian et al 2002; Du et al 2002). Therefore, these results suggest the use of HT sorghum in layer diets is not economical and should be accompanied with supplementation with more readily available energy sources. However, these results disagree with other researchers who reported a decrease in feed intake in HT diets (Attia 1998; Ali and Mahmood 2003).

The influence of tannin on egg weight is variable. Malik and Queensberry (1963) and Armanious et al (1973) reported a decrease in egg weight when maize replaced sorghum in laying hen diets. However, our findings suggest that tannins up to 2.6% DM in the diet have no detrimental effect on egg weight. These findings are consistent with other research reports (Ambulla et al 2003; Imik 2009; Kwari et al 2011).

The lower egg mass values noted in HT compared to LT dietary groups compares well with other research reports (Armonious et al 1973; Faquinello et al 2004; Kyarisiima et al 2004). These results suggest that tannins have a detrimental effect on egg mass and could be uneconomical particularly where eggs are graded.

The poorer feed conversion values noted in HT compared to LT dietary groups may be attributed to the higher feed intake and lower egg mass, due to negative effect of tannins.

The decrease of egg laying percentage in diets supplemented with molasses may be attributed to the low Metabolizable energy and almost complete absence of amino acids reported in final sugar cane molasses (Ly 1979, 1990). However, these results are inconsistent with previous research reports (Damron et al 1980; Rahman et al 1991; Valdivie 2003) which may be due to differences in type of molasses and the basal diet used (Ly 1990). The increase in egg laying percentage due to Moringa leaf meal was probably due to the effects of the higher protein availability in Moringa (Sikka and Johari 1979; Makker and Becker 1997). These finding suggest utilization of moringa leaf meal as additive in sorghum based diets for layer chickens can improve laying rate and minimize the need for supplementary synthetic amino acids. However, there are also conflicting reports on the effect of Moringa leaf meal on egg production (Abou-Elezz et al 2011).

Conclusions

- High tannin grain sorghum as a source of energy in layer diets depresses egg laying rate, egg mass production and feed conversion
- *Moringa oleifera* leaf meal is a suitable additive to grain sorghum based diets for layer chickens as it improves egg production and could eventually reduce the expenses of using synthetic amino acids.
- Final sugar cane molasses is not an appropriate additive to grain sorghum based diet for layer chickens as its use depresses egg production and feed conversion.

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CHAPTER SEVEN

7.0 PAPER V

Moringa oleifera leaf meal and molasses as additives in grain sorghum based diets; effects on egg quality and consumer preferences

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Moringa oleifera leaf meal and molasses as additives in grain sorghum based diets; effects on egg quality and consumer preferences

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Abstract

The comparative influence of *Moringa oleifera* leaf meal (MOLM) and final sugar cane molasses (SCM) in grain sorghum variety (GSV) based diets on egg quality characteristics and consumers' preferences for commercial hybrid layer chickens was studied in 2x3 factorial designs. The study used two grain sorghum varieties (GSV): high tannin sorghum (HTS) and low tannin sorghum (LTS); three supplementary strategies (SPS) of non-supplemented and 10% DM- molasses and 10% DM-MOLM supplemented GSV to make six dietary treatments. Roche yolk color score (RYS) value was higher in HTS compared to LTS. The consumers' egg aroma score index (ASI) and egg taste score index (TASI) increased in HTS. The supplementation of MOLM increased RYS and YSI. However, both supplementation of sugar cane molasses and MOLM moderately decreased ASI and GAS. It can be concluded that HTS produced moderately acceptable eggs in terms of aroma, flavor and yolk color but enhanced egg yolk pigmentation and egg flavor and aroma whereas use of LTS could produce moderately acceptable eggs in terms of yolk color, aroma and flavor. Thus, its use could require supplementation with synthetic pigments, flavor and aromatic compounds. The supplementation MOLM in GSV diets could moderately improve egg yolk colour and minimize use of synthetic pigments whereas SCM could produce poor quality egg products and its use could require supplementation with synthetic pigments. Thus, utilization of 10% of MOLM as an additive to HTS and LTS for egg quality is suggested. The utilization of SCM as additive to HTS and LTS diets is not recommended due to negative effects on egg quality.

***Key words:** pigmentation, supplementation, tannin*

Introduction

Sorghum is well adapted to marginal environment, rank fifth in global cereal hectares and is the most important grain cereal in the semi-arid tropical regions of Asia and Africa. The chemical composition of grain sorghum is comparable to maize except for being deficient in lysine, methionine and threonine (Sikka and Johari 1979; Rostagno et al 2000). Thus, grain sorghum has been used as an alternative to maize in poultry diets due to its high energy content. However, the utilization of grain sorghum in poultry has been limited due to the presence of tannins (Nelson et al 1975). Tannins in plants and fruits generally act as antioxidant and anti-pathogenic agents (Amarowicz et al 2000; Gorinstein et al 2001). However, high tannin concentration in poultry feeds may reduce nutritive value (Cherian et al 2002; Du et al 2002; Imik 2006). High levels of dietary tannins up to 2% may decrease egg quality characteristics (Potter et al 1967; Fry et al 1972; Faquinello et al 2014). Several attempts has been made to curb the effects of tannins on egg quality by using synthetic amino acids, oil, pigments and minerals (Saldanha et al 2009; Bolukbasi et al 2010; Gül et al 2012). However, these nutritional regimes have been partial and expensive. *Moringa oleifera* leaf meal (MOLM) and molasses are readily available feed ingredients in the tropics with a great potential and good source of energy, minerals, vitamins and amino acids (Mathew et al 2001; Curtin 1902). The scanty of information on the utilization of MOLM and molasses to high tannin grain sorghum (HTS) based diets for layers impelled this study with the view of evaluating the effects of MOLM and molasses on grain sorghum based commercial layers diets on egg quality characteristics and consumer egg preferences. It was hypothesized that supplementation of molasses and MOLM would improve the adverse effects of tannin in sorghum on egg quality characteristics and consumer egg quality preferences through either being with essential nutrients. Therefore, the objective of this experiment was to determine on egg quality characteristics and consumer egg preferences resulting from the use of HTS and LTS as a sole use of energy into diets of hybrid egg strain layer during initial production period and whether supplementation of molasses and MOLM could ameliorate the adverse effects of tannins.

Materials and Methods

Feed ingredients and experimental diets

The study was conducted at poultry unit of the Sokoine University of Agriculture (SUA) in the Department of Animal Science and Production (DASP), Eastern part of Tanzania. *Moringa oleifera* leaves were harvested within SUA compound, air dried under a shed to maintain their greenish colour, ground with hammer mill to produce MOLM. Grain sorghum varieties (GSV) were collected from the local markets, ground with hammer mill, passed through 2mm sieve to produce grain sorghum meal.

Molasses was obtained from Mtibwa sugar processing company. The methods of Association of Official Analytical Chemists (AOAC, 1990) were used to determine proximate composition, minerals and ash of the feed ingredients and experimental diets. Metabolizable Energy (ME/ kcal/kg) content of feed ingredients and experimental diets were estimated by prediction equations expressed by NRC (1994). MOLM, ME/Kgcal/kg was estimated by prediction equation established by Carpenter and Clegg (1956). Condensed tannins in GSV and MOLM were determined using butanol/HCL method as expressed by Nitao et al (2001). All ME_n values were converted into MJ/KgDM. All diets were formulated to meet or exceed the nutrient requirements recommended by NRC (1994) (Table 1) for layer chickens. The inclusions of GSV in experimental diets were fixed to 55%DM.

Experiment design and dietary treatments
The six dietary treatments were arranged in 2x 3 factorial design of two GSV for (HTS) and (LTS) and three supplementary strategies (SPS) for non supplemented (0%) and 10%DM-molasses and 10%DM-MOLM for supplemented GSV, making six diets designated as CTL_1, SCM_1 and MOL_1 and CTL_2, SCM_2 and MOL_2 in HTS and LTS, respectively. The name and description of experimental diets (D_1-D_6) are shown in Table 1.

Experimental procedure
Two hundred and seventy (270) commercial bovan brown hybrid layer chickens at 20th week of age were randomly allocated to dietary treatment. Each dietary treatment had 45 birds subdivided into three replicates of 15 birds each. The birds were housed deep litter pens. The experimental diets were provided from 20th to 30th week of age. The experimental diets and clean drinking water were supplied to the birds without restriction throughout the study period. Light was supplied by electric bulb and sun at night and day time respectively. Vaccination and other routine poultry management practices were carried out. Four eggs from each replicate group, 12 eggs per treatment, were randomly sampled every three weeks for nine weeks for quality evaluation of physical and internal characteristic parameters. The egg shape index was calculated and expressed as a ratio of egg length to width. The egg length was measured in centimeter (cm) along the longitudinal axis and width measured along the equatorial axis using a venire caliper. The eggshell thickness was measured using micro-meter. Egg component proportions were weighed and proportions of their weights calculated. The eggs were carefully broken, opened by hand; yolk separated, dried using blotting papers and weighed using sensitive electronic balance. Eggshell weight was taken after oven drying the eggshells at 70^0 C for 24 hr. The albumen weights were determined by the difference between the whole egg weight and that of the yolk added to the shell weight. Egg component proportions were determined based on egg weight. Egg yolk colour scores were determined using a 15 grade Roche fans (Hoffman Roche Switzerland). Egg albumen and yolk solids were determined according to the AOAC (1990) procedure. Egg yolk cholesterol concentration was determined by chod-pap method and expressed as mg/dl.
At the end of the feeding experiment, sixty (60) untrained panelists, thirty women and thirty men, were randomly selected for egg preference evaluation. Eggs were randomly selected from each treatment replicate and coded with random numbers. Each panelist was allowed to randomly select one coded egg from each dietary treatment. All sampled eggs were boiled for five minutes and served to panelist while warm. A glass of water was provided to each panelist to rinse the mouth after testing each sample. Five score Hedonic sensory test was used in egg scoring. The five scoring points were 1,2,3,4 and 5 with scoring indices of 1-20, 21-40, 41-60, 61-80 and 81-100% for dislike very much, dislike moderately, neither like or dislike, like moderately and like very much respectively. Panelists indicated the degree of likeliness for each sampled egg by choosing appropriate category which suited their attitude towards eggs. The mean score of studied parameters in each replicated group was calculated by summation of score dividing to the number of panelists. Then the mean score index was calculated by score dividing to the highest score rank x 100.

Statistical analysis

All data on physical and internal egg quality characteristic and egg quality consumers' score indices were analyzed in accordance with the 2x3 factorial designs of two GSV in combination with three SPS using General Linear Model procedure of SAS software version 9.1 for windows (2007). Values were considered significant at ($P \leq 0.05$). The Least Square Difference was used to compare means of each variable. The analytical model for studied haematological parameters and serum biochemical indices was as follows: $Y_{ijk} = \mu + V_i + S_j + (VS)_{ij} + e_{ijk}$

Where:

Y_{ijk} = Observation of k^{th} bird assigned to i^{th} GSV subjected to j^{th} SPS

μ = overall mean to all observations; I = effect of GSV based diet (HTS or LTS)

S_j = the effect of SPS based diet (CTL, SCM or MOL)

$(VS)_{ik}$ = the interaction between GSV and SPS

e_{ijk} = random error

Results

Effect of type of GSV

The overall means of physical and internal egg quality characteristics for GSV are shown in (Table 2). The egg shape index (ESI), egg shell thickness (EST), percent egg shell (PES), percent egg yolk (PEY), percent egg albumen (PEA), percent yolk solids (PYS), percent albumen solids (PAS), Roche yolk colour score (RYS) and egg cholesterol concentration (ECC) values were not influenced by type of GSV. However, the EST, PES and PEY values were moderately lower in HTS and higher in LTS dietary groups though not significant. The Roche yolk colour score (RYC) was higher in HTS and lower LTS dietary group.

The egg quality consumer' score index for GSV are shown in (Table 5). The egg yolk colour scoring index (YSI), general acceptability egg scoring index (GASI) and total egg scoring index (TSI) were not influenced by type of GSV. But, the taste score index (TASI) and aroma score index (ASI) was higher in HTS and lower LTS dietary group.

Effect of supplementation

The overall means of physical and internal quality egg quality characteristics for SPS are shown in (Table 3). The ESI, EST, PES, PEY, PEA, PYS, PAS and ECC values were not influenced by type SPS. The supplementation of SCM and MOL to GSV diets did not affect EST, PES and PEY values. The supplementation of SCM did not change whereas supplementation of MOL increased the RYS values. The interaction (Table 4) between type GSV and type SPS was noted on RYS values. RYS values were moderately higher in MOL_1 compared to MOL_2 dietary group. The supplementation SCM in HTS and LTS based diet did not change but supplementation of MOL increased RYS values.

The overall means of consumer egg quality scoring indices for SPS are shown in Table 6. The results showed that, the three SPS had no influence on TASI and TSI values. The supplementation of SCM to GSV diets decreased ASI and GAS. Moreover, the supplementation of SCM did not change whereas supplement MOL increased YSI value.

Table 1: Composition of experimental diets

Ingredient	HT	LT	HT-Mo	LT-Mo	HT-ML	LT-ML
Moringa leaf meal	-	-	-	-	11.3	11.3
Molasses	-	-	14.4	14.4	-	-
Sorghum meal (high tannin)	62.5	-	62.5	-	62.5	-
Sorghum meal (low tannin)	-	62.3	-	62.3	-	62.3
Fish meal	10.0	10.0	10.0	10.0	10.0	10.0
Sunflower seed meal	18.0	18.0	18.0	18.0	18.0	18.0
Limestone	6.50	6.80	6.50	6.80	6.50	6.80
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Bone meal	2.25	2.25	2.25	2.25	2.25	2.25
Salt	0.50	0.50	0.50	0.50	0.50	0.50
Chemical composition (%DM)						
Dry matter	88.9	90.6	87.8	87.0	89.5	88.6
Crude protein	17.0	16.7	17.9	19.3	20.5	19.8
Crude fiber	9.60	9.27	7.41	6.50	10.6	8.03
Ether extract	7.52	9.51	5.62	6.34	9.08	9.85
NFE (Starch +sugar)	33.7	34.9	39.1	39.8	39.0	39.3
Lysine	0.885	0.868	1.091	1.101	3.102	3.12
Methionine &cystine	0.5087	0.499	0.655	0.6609	1.054	1.065
Tryptophan	0.186	0.183	0.213	0.215	0.263	0.265
Tannin	2.60	1.30	2.15	1.05	2.55	1.36
ME (MJ/kgDM)	9.42	8.77	8.606	9.23	9.27	9.105

HT-.55%DM-High tannin grain sorghum based diet; LTS- 55%DM-Low tannin grain sorghum based diet; HT-Mo- 55%DM-High tannin grain sorghum+10%DM-molasses; LT-Mo- 55%DM-Low tannin grain+10%DM-molasses; HT-ML- 55%DM-High tannin grain sorghum+10%DM-Moringa leaf meal; LT-ML- 55%DM-Low tannin grain sorghum+10%DM-Moringa leaf meal.

Table 2: Physical and internal egg quality characteristics of commercial layer chickens fed grain sorghum variety based diets

	Grain sorghum variety based diets			
	HTS	LTS	SEM	Prob
Egg shape index (%)	77.2	77.0	0.972	0.635
Egg shell thickness (μ)	193	210.7	20.62	0.0915
Percent egg shell (%)	12.3	12.6	0.306	0.0990
Percent egg yolk (%)	22.5	23.7	1.24	0.0699
Percent egg albumen (%)	51.5	51.1	0.859	0.374
Percent egg yolk solids (%)	49.9	52.0	4.038	0.290
Percent egg albumen solids (%)	12.8	13.3	0.549	0.115
Roche egg yolk colour score	3.98^{a}	3.54^{b}	0.382	0.0371
Egg cholesterol concentration ((mg/dl^{-1})	353	338	58.0	0.6006

^{ab} Means in the same row with different superscripts are different (P = 0.05); SEM = Standard error of means; HTS.55%DM-high tannin grain sorghum based diet; LTS- 55%DM-low tannin grain sorghum based diet.

Table 3: Physical and internal egg quality characteristics of commercial layers fed MOLM and molasses supplemented in grain sorghum variety based diets (20-30 weeks of age)

Parameter	Supplementary diet				
	CTL	SCM	MOL	SEM	Prob
Egg shape index (%)	76.9	77.2	77.2	0.972	0.8609
Egg shell thickness (μ)	192	206	207	20.26	0.418
Percent egg shell (%)	12.3	12.4	12.6	0.306	0.273
Percent egg yolk (%)	22.7	23.0	23.7	1.24	0.345
Percent egg albumen (%)	51.4	51.3	51.2	0.549	0.947
Percent egg yolk solids (%)	51.1	51.8	49.9	4.038	0.723
Percent egg albumen solids (%)	12.9.	13.0	13.2	0.549	0.454
Roche egg yolk colour score	1.0^{b}	1.0^{b}	9.28^{a}	0.382	0.0001
Egg cholesterol concentration ((mg/dl^{-1})	293	379	365	58.0	0.584

^{ab} Means in the same row with different superscripts are different (P = 0.05); SEM = Standard error oj means; CTL.55%DM-grain sorghum+ '0' supplementation; SCM.55%DM-grain sorghum plus 10%DM' molasses supplementation; MOL.55%DM-grain sorghum plus10%DM-MOLM supplementation

Table 4: Interaction between grain sorghum variety and type of supplementary strategy on egg Roche colour score for commercial layer chickens (20-30weeks of age).

Parameter	HTS			LTS				
	CTL_1	SCM_1	MOL_1	CTL_2	SCM_2	MOL_2	SEM	Prob
Roche egg colour score	1.0^a	1.0^a	9.28^b	1.0^a	1.0^a	8.63^b	0.382	0.0215

[abc] *Means in the same row with different superscripts are different ($P = 0.05$);* SED = *Standard error of means; CTL_1-55%DM- high tannin grain sorghum + '0' supplementation;CTL_2 -55%DM- low tannin grain sorghum+ '0' supplementation; SCM_1.55%DM- high tannin grain sorghum plus 10%DM' molasses supplementation; SCM_2.55%DM- low tannin grain sorghum plus 10%DM' molasses supplementation; MOL_1.55%DM- high tannin grain sorghum plus10%DM-MOLM supplementation; MOL_2.55%DM- low tannin grain sorghum plus10%DM-MOLM supplementation.*

Table 5: Consumer egg quality scoring indices of commercial layer chickens fed two types of GSV based diets

Parameter (%)	Grain sorghum variety based diets			
	HTS	LTS	SEM	Prob
Egg taste score index	73.6^{a}	67.3^{b}	5.34	0.0329
Egg aroma score index	64.2^{a}	59.6^{b}	3.96	0.030
Egg yolk colour score index	63.1	63.8	8.96	0.878
Egg general acceptability score index	65.11	62.00	5.39	0.249
Egg total consumer score index	50.8	48.9	2.42	0.128

^{ab} *Means in the same row with different superscripts are different (P = 0.05)*; SEM = Standard error of means; *HTS.55%DM-high tannin grain sorghum based diet; LTS- 55%DM-low tannin grain sorghum based diet. Scoring index: 1-20%-dislike very much; 21-40%-dislike moderately; 41-60%- neither like nor dislike: 61-80- like moderately and 81-100%-like very much*

Table 6: Consumer egg scoring indices for commercial layers chickens fed MOLM and molasses supplemented in grain sorghum variety based diets (20-30 weeks of age)

Parameter	Supplementary diet				
	CTL	SCM	MOL	SED	Prob
Egg taste score index (%)	73.0	68.3	70.0	5.34	0.347
Egg aroma score index (%)	67.3^{a}	58.3^{b}	60.0^{b}	3.96	0.0063
Egg yolk color score index (%)	53.7^{b}	56.7^{b}	80.0^{a}	8.96	0.0009
Egg general acceptability score index (%)	67.3^{a}	63.3^{b}	60.0^{bc}	5.39	0.1091
Egg total consumer score index (%)	51.2	48.3	50.0	2.42	0.175

^{ab} *Means in the same row with different superscripts are different (P = 0.05); SED = Standard error of difference; CTL.55%DM-grain sorghum+ '0' supplementation; SCM.55%DM-grain sorghum plus 10%DM' molasses supplementation; MOL.55%DM-grain sorghum plus10%DM-MOLM supplementation. . Scoring index: 1-20%-dislike very much; 21-40%-dislike moderately; 41-60%- neither like nor dislike: 61-80- like moderately and 81-100%-like very much.*

Discussion

In the present study PEA, PYS and PAS values did not differ significantly in HTS and LTS dietary groups. These results were in conflict with other previous studies (Ali and Mahmood 2003; Ebadi et al 2005; Imik 2009) that showed negative effect of tannins on albumen and yolk solids. These results probably imply that dietary tannin in the present study was not high enough to affect those parameters. These findings suggest that it is possible to utilize HTS with dietary tannin up to 2.6% of the diet DM as leucocynidin equivalent without detrimental effect on internal egg characteristics.

ESI, EST and PES values were comparable in HTS and LTS dietary groups in the present study. These findings are in agreement with previous research reports (Faquinello et al 2004; Imik et al 2006). However, they were in contrast with those of Ali and Mahmood (2003) that reported a decrease in egg shell thickness and egg shell strength in birds fed HTS based diets. Tannin intake is negatively correlated with egg shell thickness (Ebadi et al 2005). Tannins may reduce availability of minerals and change cation- anion balance in blood and availability of HCO_3 and eventually reduce egg shell thickness (Sell et al 1983, 1984). Tannins may also affect bio-availability of calcium (Mehansho et al 1987; Chang et al 1993) and reduce egg shell thickness. These findings may suggest that dietary tannin in the present study was not high enough to negatively affect those parameters or a high level of dietary protein probably offsets the negative effects of tannins (Sell et al 1983). These findings suggest that there is a possibility of utilizing HTS based diet with dietary tannins up to 2.6% in DM as leucocynidin equivalent in layer chickens without getting detrimental effects on physical egg shell characteristics and egg shell quality if diets are supplemented with adequate calcium or have high protein content.

In the present study RYS values were higher in HTS and lower in LTS dietary groups indicating presence of more pigments in HTS than LTS. These results are in agreement with other previous research reports (Çabuk et al 2004). Polyphenolic compounds such as condensed tannins have been reported to increase deposition yellow yolk pigments and decrease malonaldehyde formation in the yolk. Also, anti-oxidants such as condensed tannins may cause excessive deposition of pigments in the egg yolk (Loetscher et al 2003;_Chukwuka et al 2011) and that minimizes losses of oxycarotenoids which occurs due oxidation particularly during storage (Seemann, 2000). These findings imply that lower RYS values noted in LTS treatment groups probably were associated with losses of pigments due to oxidation whereas higher RYS values observed in HTS could be due to excessive deposition of pigments by tannins that offset losses of pigments during storage. However, these findings are in conflict with other research reports (Ali and Mahmood 2003; Faquinello et al 2004) that showed a decrease of pigments in egg yolk with increase of tannin intake. The conflicting results might be associated with differences in storage conditions and level of pigments in grain sorghum.

The effect of tannin on egg cholesterol content is variable and dose dependent. In the present study, egg cholesterol concentration values amongst HTS and LTS dietary group were similar. These findings suggest that the effect of tannins on egg cholesterol were not evident. These results disagree with other research reports

(Stensvold et al 1992; Ali and Mahmood 2003; Oh et al 2013). The reasons for conflicting results in the present study may be associated with differences in dietary tannin content. These findings suggest that dietary tannins up to 2.6% of DM as leucocynidin equivalent were not high enough to reduce egg cholesterol concentration.

Acceptability of egg yolk color by consumers for eggs from HTS and LTS diets was moderate. These findings may be related to the pale egg yolk products noted in LTS and HTS dietary groups due to absence of pigments in GSV. Consumers perceive that eggs with pale yolks are not attractive and could be poor in nutritive value. Yolk colour is a very important parameter by which consumers judge the quality of eggs (Jacob et al 2000) and is an important egg quality attribute for egg marketing in different countries. These findings suggest that utilization of HTS and LTS as a main source of energy for layer chickens could produce undesirable egg products. The visual yolk color liked by consumers is the result of the deposition and coloring capacity of oxy-carotenoids, called xanthophylls, in the egg yolk (Coutts and Wilson 1990). Sources of xanthophylls can be natural or synthetic. Thus, the utilization of GSV as a main source of energy for layer chickens should be supplemented either by natural or synthetic pigments to improve egg acceptability by consumers.

Egg flavor acceptability by consumers for eggs from HTS and LTS diets was moderate. These findings suggest that the presence of phenolic compounds other than tannins may affect sensory characteristics and nutritional quality of grain sorghum and its subsequent products (Hahn et al 1984). These compounds alone or in combination with other compounds give rise to sensory characteristic that impart astringency, bitter or sour tastes (Maga and Lorenz 1973) which may affect overall acceptability of eggs. However, higher egg taste scoring index was noted in HTS whereas lower taste scoring index was observed in LTS diets, and may have been due to differences in tannin. The taste of bitterness and the tactile sensation of astringency are elicited primarily by flavanol polymers (proanthocyanidins or condensed tannins). Variations in proanthocyanidin composition, such as polymer size, extent of galloylation, and formation of derivatives, affect both bitterness and astringency. The social influence could be the reason for higher egg taste scoring index in HTS (Lesschaeve and Noble 2005). These findings suggest that utilization of GSV as a main source of energy could require supplementation with flavour rich compounds to increase acceptability of eggs.

Aroma acceptability by consumers for eggs from HTS and LTS diets was indecisive. These findings may be attributed to loss of desirable aromatic compounds in HTS and LTS during storage. However, higher egg aroma acceptability noted in HTS diets and lower egg aroma acceptability observed in LTS diets suggest the presence of higher aromatic compounds (ashy or smokey) in HTS due to tannin effects and lower aromatic compounds in LTS due to oxidation during storage. Tannins as natural antioxidants enable retention of aromatic compounds from the action of oxidasic enzymes, such as laccase and free radicals that are formed from the oxidation of polyphenolic molecules. These findings suggest utilization of HTS and LTS could require supplementation of aromatic compounds to improve aroma acceptability in eggs.

The supplementation of SCM and MOLM to GSV based diet slightly increased ESI, EST, PEA and PEY values indicating inadequacy of nutrients particularly minerals in GSV. Thus, supplementation with molasses and MOLM compensated deficient nutrients in GSV to optimize egg shape index, egg shell thickness, egg albumen and yolk weight values. These findings suggest use of SCM and MOLM as additives to GSV could slightly compensate deficient minerals particularly calcium and minimize cost of using synthetic minerals. These results agree with other previous studies (Olugbemi et al 2010; Ebenebe et al 2013). However, the supplementation of MOLM and SCM to GSV had no influence on PAS and PYS values suggesting adequacy of nutrients particularly proteins and amino acids responsible for development of albumen and egg yolk solids in GSV. These findings suggest utilization of SCM and additives can improve physical characteristics of eggs.

Supplementation with SCM to the GSV based diet did not change RYS whereas supplementation with MOLM increased RYS values. These results may be attributed to absence of xanthophylls and carotenoids in SCM (Kaijage et al 2014) and presence of xanthophylls which are characterized by the presence of hydroxyl groups and carotenoid pigments in MOLM (Fuglie 1999). Colour is the most important quality attribute of egg yolk. Consumer's preferences for yellow yolk colour have been linked with high nutritional value and attractiveness of eggs (Wells 1968). Therefore, these results suggest that utilization of SCM as additive to GSV based diets for poultry cannot improve desirability of egg products in the egg marketing industry and its utilization will necessitate use of synthetic pigments to produce colored yolk products to improve acceptability. These findings suggest that utilization of MOLM as additive to GSV can improve the egg yolk colour desirable to consumers; and that its use in the poultry industry could promote the use of natural pigments in situations where synthetic pigments are expensive or their availability is restricted. These results concur to other previous research reports (Kaijage 2003; Olugbemi et al 2010; Abou-Elezz 2011).

However, the interaction between GSV and SPS on RYS observed in the present study suggests differences between HTS and LTS in he amounts of pigments. The HTS had moderately higher whereas LTS had lower RYS suggesting higher concentrations of pigments in HTS and lower concentrations in LTS. These findings support other studies that tannins can enhance deposition or prevent oxidation of pigments (Faquinello et al 2004). Therefore, HTS could enhance absorption of natural and synthetic pigments and minimize loses of pigments particularly during storage, and eventually produce desirable and acceptable egg yolk colour.

In the present study, supplementation of SCM and MOLM to GSV had no influence on ECC values. These findings disagree with other previous research studies (Ghasi et al 2000; Olugbemi et al 2010; Lala et al 2012) that showed SCM and MOLM to have hypocholesterolemic properties. The hypocholesterolemic nature of MOLM may be partly explained by high fiber content in MOLM that block intestinal cholesterol absorption (Lansky et al 1993) or presence of bioactive phytochemicals, the phytosterols such as β-sito-sterols, campe-sterols, stigma-sterols and avenasterol in MOLM that lower plasma concentration of low density lipoproteins (LDL), cholesterol (Ghasi et al 2000; Mbikay 2012) and ultimately lower cholesterol concentration in eggs. Sterols are also reported to inhibit cholesterol production in the liver. Further, the hypocholesterolemic mechanisms of SCM are associated with a series of n-alkenes (C_{23}-C_{33}) and ethyl and methyl esters of fatty acids (mainly oleate and palmamitate) of phytosterols (stigma-sterol, campestrols, and β-sito-sterols), free fatty acids and triglycerides and keto-steroid derivatives contained in molasses which could inhibit intestinal cholesterol absorption and thereby lower plasma total LDL cholesterol levels (Ghasi et al 2000) and ultimately could lower cholesterol in eggs. The reasons for conflicting results in the present study with other previous studies are not clear but could be due to differences in inclusion of SCM and MOLM. These findings suggest utilization of SCM and MOLM as additives to GSV based diets has no influence on egg cholesterol concentration.

The present study showed that flavor of eggs from three types of SPS diets were moderately accepted by consumers. The sugary flavor from compounds such as acetic acid, valeric acid, benzoic acids and syringic acids reported in sugar cane molasses (Takesh et al 1967) and pungent flavor caused by phytochemical-2-pentatione reported in moringa leaves were not evident in eggs. These findings suggest that supplementation of SCM and MOLM as additives to GSV based diet did not improve consumer acceptability for egg flavor.

Acceptability of aroma in eggs from the three types of SPS diets was uncertain. These findings suggest that supplementation of SCM and SCM had no influence on egg aroma. However, the decrease in aroma acceptability in SCM and MOL compared to CTL was associated with slightly strong fruity aroma reported in molasses and moringa leaves (Pinnott et al 2006). Therefore, the use of 10% SCM and MOLM as additives to GSV based diets may negatively affect acceptability of egg aroma.

The supplementation of SCM to GSV based diet did not change customer preference whereas supplementation with MOLM increased preference for egg yolk color. These findings were attributed to pale egg yolk noted in SCM diets due to lack of pigments and yellow egg yolk observed in MOLM due to presence of pigments. Consumers prefer pigmented egg yolk such as yellow or deep yellow (Faquinello et al 2004). The pigmented egg yolk is associated with high nutritive value. Therefore use of molasses as additive to GSV based diet could produce undesirable egg products. Therefore, its utilization may require supplementation of synthetic pigments for production of coloured yolk egg products preferred by consumers. However, use of moringa leaf meal as additive to GSV improves egg yolk colour preference by consumers and could minimize use of synthetic pigments. These results agree with previous research findings (Kaijage 2003; Olugbemi 2010).

Conclusions

- Utilization of HTS as main source of energy for layer chickens could produce moderately acceptable egg products in terms of yolk colour, flavor and aroma and could enhance absorption of pigments and minimize losses of pigments, flavor and aromatic compounds. However, there is probably a need for additional supplements to provide desirable pigments, flavor and aromatic compounds to increase egg acceptability.

- The utilization of LTS could produce moderately acceptable egg products in terms of yolk colour, flavor and aroma. Its use for layer chickens, however, could require supplements to provide desirable pigments, flavor and aromatic compounds to increase egg acceptability.
- The supplementation of final sugar cane molasses as an additive to HTS and LTS could worsen egg yolk colour score and egg yolk colour acceptability. Thus, its utilization for improving feeding value for poultry cannot be economical as this would require supplementation with synthetic pigments to increase egg acceptability.
- The supplementation of MOLM in HTS and LTS based diet can improve egg yolk colour score and increase egg yolk acceptability by consumers. Thus, its utilization could be economical as it reduces need for synthetic pigments.

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CHAPTER EIGHT

8.0 DISCUSSION, CONLUSIONS and RECOMMENDATIONS

8.1 General Discussion

The overall objective of the study was to improve feeding value of Tanzanian GSV for poultry by evaluating its nutritive value and the effectiveness of MOLM and SCM in improving their feeding values. In the present study, the Tanzanian commercial GSV were identified and their nutritive values were determined. Thereafter, two experiments were conducted to evaluate if SCM or MOLM could be strategic option for improvement of GSV feeding value for poultry. The first experimental study compared effects of SCM and MOLM supplementation in GSV based diet on performance, hematology and serum bio-metabolic indices of growing chicks. The second experimental study compared the effects of final sugar cane molasses and MOLM supplementation in GSV based diets on egg production performance, egg quality characteristics and consumers' egg quality preferences. The research outputs this thesis is discussed as follows:-

A total of twelve Tanzanian commercial GSV, mostly from Singida and Dodoma, were identified and were predominantly landraces. Most of them had white seed coat color. However, the study did not find any significant correlation between coat colour and level of tannin in GSV. The GSV were generally high in dry matter, energy and starch values that could indicate their potential to contribute substantial amount of feed components, in particular energy, in poultry diets. The GSV were however, generally low in micro/ macro minerals, amino acids, and high in contaminants, crude fiber and condensed tannins. These features in a way may compromise the feeding value and thus limit their full utilization for poultry. This shows that in order to have optimum utilization of Tanzanian GSV in poultry diets supplementation of minerals, amino acids or strategic feeding options are needed so as to limit the anticipated negative effects due to condensed tannins and nutrient deficiency.

The insignificant differences between birds fed LTS based diets and those birds fed HTS based diets on growth parameters observed in the present study showed that it was possible to use HTS as main source of energy without causing any adverse effect on growth performance in chicks. These results imply that dietary tannins up to 2.6%DM as leucocynidin equivalent found in HTS diets have no effect on growth performance of chicks. The present findings revealed that growing chicks fed LTS and HTS based diets had pale body parts which was probably due to the lack of pigments reported in GSV. Additionally, it has been reported that grain sorghum has the potential to produce undesirable coloured poultry products. Leg abnormalities were noted in birds fed GSV based diets but leg abnormality index was higher in LTS dietary group. These results signified the deficiency of minerals particularly phosphorous in HTS and LTS probably caused by the anti-nutritional effects of tannins in HTS and phytates in LTS. In the present study FCR was better for LTS based diets compared to HTS diets in growing chicks and this was probably due to energy and protein inadequacy in HTS based diets caused by tannins.

The lower lymphocyte and oesinophils ratios and higher monocyte ratios observed in birds fed LTS based diets was an indication of adequate nutrients in terms of energy, proteins and amino acids in LTS. However, the increase of lymphocyte and oesinophil ratios and decrease of monocyte ratio observed in birds fed HTS diets for growing chickens was an indication of nutrient deficiency such as energy, proteins and amino acids due to negative effects of tannins in HTS. These finding shows that LTS could maintain cell mediated immunity in growing chicks whereas HTS could lead to nutritional stress and disrupt cell mediated immunity.

The present findings showed that birds fed LTS had higher egg production whereas egg production was lower for birds fed HTS based diets. These results reflected adequacy and inadequacy of nutrients responsible for egg production in LTS and HTS based diets respectively. These results showed that using LTS as a main source of energy for layer chickens could optimize nutrients for egg production whereas HTS could reduce availability of nutrients for egg production. Higher feed conversion ratio observed in the present study in birds fed HTS based diets compared to those birds fed LTS based diets was suggesting energy inadequacy in HTS and adequacy in LTS. These findings were attributed to the negative effects of tannins in HTS. These results showed that utilization of LTS as a source of energy for layer chickens could optimize energy for egg production whereas HTS could reduce energy for egg production. The present study showed that the acceptability of eggs in terms yolk colour, aroma and flavor by consumers were moderate in HTS and LTS diets suggesting absence or loss of pigments and desirable flavor and aromatic compounds during storage in GSV. However, higher RYS, TAS and ASI in HTS diets and lower in LTS diets were suggesting loss of pigments and flavor and aromatic compounds in LTS and ability of tannins to increase absorption of pigments and maintain flavor and aromatic compounds particularly after being stored for a long period.

The present findings showed that addition of SCM in LTS and HTS based diets increased prevalence of LAI for growing chicks suggesting deficiency of mineral particularly phosphorous in sugar cane molasses diets. On the other hand addition of MOLM prevented occurrence of LAI signifying the adequacy of minerals responsible for bone development in Moringa leaf meal diets. These results imply that utilization of sugar cane molasses as additive in GSV based diet could negatively affect utilization of minerals in growing chicks, probably due to increased rate of passage in the gastro-intestinal tract. Thus, sugar cane molasses based diets could be improved by supplementing synthetic minerals for growing chicks. The results further showed that inclusion of Moringa leaf meal in GSV based diets could alleviate the negative effect of tannins/ phytates and compensate for deficiency of minerals in GSV may be due to the presence substantial amount of minerals reported in Moringa leaves.

Addition of SCM in LTS and HTS based diets increased paleness of the bird body parts probably due to inadequate pigments in SCM whereas addition of MOLM led to yellowing of the bird's body parts. These findings clearly showed that sugar cane molasses lacked pigments and could lead to production of undesirable colored egg yolk. Thus, its use in poultry feeding may require supplementation of synthetic pigments where coloured poultry products are preferred. Furthermore, the findings were showed that MOLM contained substantial amount of pigments and led to the production of desired colored egg yolk.

The slight improvement in feed conversion ratio for growing chickens observed when 10% SCM and MOLM were added to HTS based diets was due to an increase in the availability of energy and other nutrients. However, the improvement was not as high as expected probably due to increased rate of passage of feed in the GIT in SCM/HTS based diets and high crude fiber levels in MOLM/HTS based diets. Thus these findings indicate that lower levels of SCM and MOLM in HTS based diets might be more beneficial. However, inclusion of SCM and MOLM in HTS based diets led to reduced lymphocyte and oesinophils and increased monocyte ratios for growing chicks whereas the opposite was observed for LTS based diets. These observations were attributed to the reduction of negative effects of tannins by SCM and MOLM in HTS based diets. This eventually might have increased energy and amino acids availability and thereby improving the nutritional status and cell mediated immunity for growing chicks. The addition of SCM to LTS based diets depressed lymphocyte ratio and increased monocyte ratio whereas addition of MOLM did not change lymphocyte and monocyte ratio suggesting nutritional adequacy in LTS. Thus, utilization of SCM and MOLM as a strategic option for improvement of LTS could not be necessary.

The decrease in egg production observed for layers when SCM was added to HTS based diets suggest that SCM impaired utilization of nutrients responsible for egg production probably increased rate of passage of feeds in gastro-intestinal tract. Additionally, the inclusion of SCM in LTS and HTS diets showed low RYS and YSI suggesting inadequacy of pigments in SCM based diets whereas addition of MOLM increased RYS and YSI suggesting adequate amount of pigments in MOLM diets. Thus, utilization of SCM may require supplementation of synthetic minerals in commercial egg production for improvement of RYS and YSI. On the other hand the adequate pigments in MOLM led to production of desirable yellow coloured egg yolk products.

8.2 General Conclusions

The research findings of this thesis can be generally concluded as follows:-

Tanzanian commercial GSV have high potential to be used in poultry feeding but their fully utilization could be limited by the presence of anti-nutritional factors such as condensed tannins and being deficient in macro and micro minerals and quality proteins.

The HTS has lower feeding value than LTS in poultry feeding due to the negative effects of tannins which impair utilization of carbohydrate component, minerals and amino acids and being deficient in pigments. It leads to poor feed conversion, leg abnormality incidences, disruption of cell mediated immunity and nutritional stress in chicks. Moreover, has poor feed conversion, depress egg production and impair physical egg characteristics in layer chickens though they can produce more desirable eggs in terms of aroma and flavour if stored for long time.

The LTS have higher feeding value than HTS in poultry feeding due to being adequate in available energy, proteins and essential amino acids. It use leads to better feed conversion, higher cell mediated immunity and nutritional status in growing chicks. In addition, has higher egg production and better feed conversion in layer chickens though can cause leg abnormalities in chicks if it contains high phytic acid and unpleasant egg aroma and flavour when stored for long time.

The use of MOLM additive to HTS based diets improve feeding value of HTS in poultry due to its ability to alleviate the negative effects of tannins by compensating deficient minerals, energy, amino acids, proteins and pigments in HTS. It use in growing chicks prevents leg abnormalities; improves feed conversion, cell mediated immunity and nutritional status. It also, improves egg production, feed conversion, physical and internal egg characteristics and desirability of eggs in terms of yolk colour in layer chickens. Moreover, the utilization of MOLM additive to LTS based diets improve feeding value of LTS for poultry due to its ability to alleviate the negative effect of phytic acids and compensate deficient minerals and pigments in LTS. This leads to control of leg abnormality incidences in chicks and improves physical egg characteristics and preference of eggs in terms of yolk colour.

The use of SCM additive to HTS and LTS based diets worsens feeding value for poultry due to the negative effect of SCM on utilization of minerals, amino acids, carbohydrate component and proteins. Its utilization in chicks aggravates leg abnormalities incidences and paleness of body parts and worsens egg production, feed conversion, physical egg characteristics and preference of eggs in terms of yolk colour and flavour.

8.3 General Recommendations

Based on the findings obtained from this study, the following recommendations are suggested:-

Partial utilization of Tanzania GSV as a source of energy in poultry feeding is suggested but their fully use should be accompanied by supplementation of synthetic minerals, pigments and amino acids or strategic feeding options to curb noted deficient nutrients and negative effects of tannins.

Moderate use of HTS as a source of energy in poultry diets is suggested but their fully use should be accompanied by supplementation of synthetic minerals, pigments and amino acids or strategic feeding options to curb noted deficient nutrients and negative effects of tannins. However, fully utilization of LTS as main source of energy in poultry' diets is highly recommended but should be supplemented with synthetic pigments and minerals for production health birds and desirable eggs.

Addition of 10%DM of MOLM to GSV based diets for improving feeding value of HTS and LTS for poultry is recommended. However, addition of 10%DM of SCM to GSV based diets for improving HTS and LTS for poultry is not suggested.

The current study focused on evaluating nutritive value of commercial grain sorghum varieties for poultry. Further, research is required to evaluate nutritive value of grain sorghum available in rural areas of Tanzania. This will enable screening suitable and unsuitable GSV for poultry for promotion and improvement respectively.

The present study evaluated 10%DM -MOLM as additive to GSV based diet for poultry. Future research is required to evaluate different MOLM additive levels to GSV based diets in different dietary tannin levels. This information will establish level of MOLM required improving feeding value of GSV with different tannin content for poultry.

Future research should focus on evaluation of economics of MOLM additive to GSV based for poultry. This information will enable understanding if it is beneficial or not beneficial to utilize MOLM as a strategic option for improvement feeding value of HTS and LTS for poultry.