PERFOMANCE OF EXOTIC CHICKEN FED INSECT-BASED DIETS: EFFECT ON LAYING CAPACITY, EGG CHARACTERISTICS AND ECONOMIC RETURNS

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DECLARATION AND RECOMMENDATION

Declaration

I hereby declare that this thesis is my original work and that no part of it has been presented for a degree in this or any other University. However, work of other researchers and authors which serve as sources of information are duly acknowledged.

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Recommendation

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DEDICATION

I dedicate this work to my dear parents, my sisters and brother, my nephews and my daughter Angel Cheryl for their continued support.

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ABSTRACT

Increasing demand for soybean meal and fishmeal for use in formulating animal feeds has led to increased prices and moreover their availability in future might be limited. Insect rearing can be part of the solution because they take a short period to mature, require small space and fewer resources to start. This study was designed to evaluate the effect of substituting fishmeal (FM) with black soldier fly larvae (BSFLM) at different inclusion levels, as a protein source in exotic chick, grower, and layers diets. The following parameters were measured, weight gain, feed intake, feed conversion ratio (FCR), laying percentage, egg characteristics and economic implication. A total of 250-day old Isa Brown layer birds were used for this experimental trial that ran for 45 weeks. Diets were formulated based on five different inclusion levels of BSFLM to replace FM (0%, 25%, 50%, 75% and 100%). Chicks were randomly distributed into five treatments each with five birds per replicate and nine replications, while a total of seventy five (75) layer hens were randomly placed in different cages each with 5 hens replicated 3 times and offered five different treatments. Data on weight gain, feed intake, egg characteristics, laying percentage, FCR and economic implication were analyzed using one way analysis of variance (ANOVA) in SAS version 9.00 (2002) with completely randomized design (CRD) model. The significance between treatment means was tested at statistical significance level of 5% and separated using Tukey's test. The results showed that most parameters were significantly ($P<0.001$) different in chick's experiment. However, there were no significant ($P>0.05$) difference recorded for grower birds' parameters. The study also demonstrated that laying capacity was significantly ($P<0.001$) different while egg characteristic was not significantly ($P>0.05$) different. Conventional layers diet (T1) was most expensive (Kes 45/kg) while T5 was the cheapest (Kes 42/kg). In conclusion, high performance can be achieved at lower inclusion levels of BSFLM up to 50% in chicks' diet and up to 75% in grower birds and layer hens' diets. Thus, BSFL is a suitable alternative to FM in exotic birds' diet.

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LIST OF ABBREVIATIONS AND ACRONYMS

Ad lib.	*Ad libitum* (Free choice)
ADFI	Average daily feed intake
ADG	Average daily gain
ANOVA	Analysis of variance
AOAC	Association of Official Analytical Chemists
BSF	Black soldier fly
BSFL	Black soldier fly larvae
BSFLM	Black soldier fly larvae meal
CBR	Cost benefit ratio
CRD	Completely randomized design
DCP	Di-calcium phosphate
FCE	Feed conversion efficiency
FCR	Feed conversion ratio
FM	Fishmeal
FRS	Free range system
GDP	Gross domestic product
HDEP	Hen day egg production
HU	Haugh unit
ICIPE	International Centre of Insects Physiology and Ecology
KALRO	Kenya Agriculture and Livestock Research Organization
Kcal	Kilo calories
KES	Kenya shillings
LSD	Least square difference
ME	Metabolizable energy
MoALF	Ministry of Agriculture Livestock and Fisheries
MoLD	Ministry of Livestock Development
MT	Metric Tonne
MJ	Mega joule
NCAP	Non- conventional animal protein
NRC	National Research Council
RoI	Return on investment
SAS	Statistical Analysis System

CHAPTER ONE
INTRODUCTION

1.1 Background Information

Agricultural sector is dominant in the Kenyan economy and accounts for approximately 25% of the country's Gross Domestic Product (GDP). Livestock contributes about 12% of Kenya's GDP, 47% to the agricultural GDP and employs 50% of agricultural labour force (MoALF, 2012). Poultry has recorded the highest livestock population of 31.8 million birds out of which 25.7 million are indigenous and 6.1 million of commercial type (GoK, 2010). Egg production globally has increased by 30.2 million tons in 1993, reaching 68.3 million tons in 2013 (Leeson, 2010). The United Nations' Food and Agriculture Organization has estimated that egg production will increase to 89 million tons in 2030 (faostat3.fao.org/home/E; Marono et al., 2017). Poultry sector is a fast-growing industry in Kenya driven by increased demand for poultry products hence leading to increased production. Chicken eggs and meat contribute to the protein nutrition of various households in the country. About 90% of the small-scale farmers in Kenya rear poultry, majority of which are indigenous chicken and exotic chicken breeds (Gichohi & Maina, 1992; King'ori et al., 2010). Sale of poultry products leads to increased and diversified revenue in the livestock sector, creates employment and promotes overall economic development (King'ori et al., 2010). Relative to other livestock species, chicken production has the advantages of having quick returns to investment and relatively simple management practices with numerous market outlets for products. Sale of products especially eggs in low value units make chicken products affordable to the low-income families especially in rural areas (King'ori et al., 2010).

High feed cost in poultry production is the major challenge faced by international as well as local industries due to competition for feed materials shared by animals and humans, hence typically low quality raw materials are mostly used in poultry feed production (Abidin et al., 2011). Current food insecurity in most developing countries and challenges of feeding over 9 billion people by 2050, has raised the need for more research to solve this problem. This is aimed at identifying other alternative sources of protein that can be used as food and feed other than, the commonly used animal protein. Also, as a result of increased income, urbanization, environment and nutritional concerns, the global food system is undergoing a profound change. There has been a major shift in human diets with increased consumption of animal products and this is likely to continue in the coming decades (King'ori et al., 2010).

Fishmeal (*Rastrineobola argentea*) is the most preferred protein source in feed formulation used by feed millers in Kenya (Munguti et al., 2012). In the year 2005, fishmeal

(FM) was the most cost-effective protein source with price ranging between Kes 40,000 and Kes 90, 000 per metric tonne (MT). However, in 2006 the prices increased to over Kes 150,000 per MT due to increased demand for fish use in feed and food. As a result, there was increased pressure to seek alternatives to replace it in animal feeds (Hardy, 2010). The search for alternative protein sources to be used as partial or complete substitute to fishmeal in chicken feed formulation, has been long and expensive. The demand for animal feed grade fish and FM significantly exceeds availability. Availability of hazards such as pathogenic salmonella contamination rise in FM, has led to most chicken feeds (especially breeder rations) to be formulated without supplementation of fish ingredient.

Insect rearing could be one of the major ways to enhance food and feed security by replacing FM use in feed formulation (Van Huis et al., 2013). Insects have various advantages which include, fast growth and reproduction rate, have high feed conversion efficiency (since they are cold blooded) and can be reared on bio-waste streams. Insects can be used both as human and livestock food/feed for the rapidly growing population (Makkar et al., 2014). Nevertheless, use of insects in animal feeds has been less studied and they exhibit great potential for development as standard ingredient in animal feed. The objective of this study was to determine the best inclusion levels of black soldier fly larvae (BSFL) in exotic chicken diet on growth performance of exotic chicks and grower pullets, egg laying capacity, egg characteristics of exotic layers and economic returns.

1.2 Statement of the Problem

Fishmeal (*Rastrineobola argentea*) as the main source of animal protein has an excellent amino acid profile, high digestible energy, crude protein (CP), minerals, vitamins, crude fat and omega-3 and omega-6 essential fatty acids. However, the amount of wild caught fish has been decreasing for years due to water pollution and high salinity levels caused by human practices. Also, incidences of salmonellosis have been on the rise thus reducing the reliance on FM as a protein source in chicken feeds. All these factors have led to high competition for FM use by man and livestock industry. As a result, this ingredient is increasingly becoming scarce and expensive for use in the poultry industry. This is due to its rapid growth leading to increased demand for chicken feeds despite the already inadequate supply of FM, which is an essential component in chicken feed formulation. The available FM cannot sustainably maintain the volumes required in processing of chicken diets, other livestock meal and for human consumption. There is need therefore, to give more consideration to alternative protein sources which also contain high omega-3 and omega-6 essential fatty acids that can be effectively used as FM replacement in chicken feed formulation.

1.3 Objectives

1.3.1 Broad Objective

To contribute to food security and better livelihoods through enhanced poultry production by substituting fishmeal (*Rastrineobola argentea*) with black soldier fly larvae meal in exotic chicken diets.

1.3.2 Specific Objectives

(i) To determine the optimal inclusion levels of BSFLM on performance (daily weight gain, daily feed intake, overall weight gain and feed conversion ratio) of exotic chicks.

(ii) To determine the optimal inclusion levels of BSFLM on performance of exotic grower pullets.

(iii) To determine the best inclusion level of BSFLM on daily weight gain, daily feed intake, feed conversion ratio, laying capacity and egg quality of exotic layer hens.

(iv) To determine the cost implication of substituting FM with BSFLM in exotic layer hens.

1.4 Hypotheses

(i) Performance of exotic chicks was not significantly different when fed diets with different inclusion levels of BSFLM.

(ii) Performance of exotic grower pullets was not significantly different when fed diets with different inclusion levels of BSFLM.

(iii) Laying capacity, egg quality, daily weight gain, daily feed intake and feed conversion ratio of exotic hens was not significantly different when they were offered different levels of BSFLM in their diets.

(iv) Cost implication of substituting FM with BSFLM in exotic layer hens was not significantly different.

1.5 Justification

Chicken have fast growth rate, minimum space requirement and effective feed utilization. Increased competition for FM between man and livestock has raised the need to identify other quality and affordable sources of protein for use in chicken feed. Black soldier fly larvae contain 42-50% crude protein (CP) and 35% fats therefore, it can be used as a FM substitute in exotic chicken diet. They can grow on a number of organic substrates thus incorporating organic waste into production systems, leading to reduced environmental pollution (Diener et al., 2009). Insects possess chitin which is found in the exoskeleton and is a non-toxic and bio-degradable linear polymer which has positive effect on the immune response of an animal (Veldkamp et al., 2012). There is minimum documented research on the effects of BSFLM on growth performance and optimal inclusion level in exotic layers diet. This informed the current study to determine the effect of BSFLM based diet on growth performance, laying percentage and egg quality of exotic layer chicken.

1.6 The Scope and Limitations of the Study

The study mainly involved substituting FM with BSFLM at different inclusion levels in the diet of exotic chicken and was conducted in Nakuru County for a period of 45 weeks. Problems encountered include increased cost of feed ingredients due to weather changes making feed formulation hectic and expensive, increased lameness incidences in chicks and high diarrhea during the grower phase. There was also, incidences of egg eating during the laying stage which was minimized by addition of calcium in feeds and early egg collection from the laying nests. The current research findings on different inclusion levels of BSFLM are limited to exotic chicken from chicks to layers stage only.

CHAPTER TWO
LITERATURE REVIEW

2.1 Livestock Production and its Role in Food Security

Livestock sector is one of the fastest growing agricultural sub-sectors in developing countries. Its contribution to agricultural GDP is already 33% and is fast increasing. It also contributes one-third of the protein that people consume. This growth is driven by the rapidly increasing demand for livestock products, which has been triggered by population growth, urbanization and increasing income in developing countries (Delgado, 2005; Thornton, 2010). Rearing of livestock plays an important role in enabling smallholder farmers to have resilient livelihoods and to avoid both food insecurity and poverty, as livestock can contribute up to 33% of household income. Overall, 75% of rural people and 25% of urban people depend on livestock for their livelihoods (Grace, 2012; Nabarro & Wannous, 2014).

2.2 Poultry Production in Kenya

Kenya has an estimated 29 million poultry population, out of which about 28.7 million (98%) are indigenous chicken. Although other poultry species are increasingly becoming important, they are comparatively few (2%) that include ducks, turkeys, pigeons, ostriches, guinea fowls and quails. About 22 million (77%) are indigenous or crosses with exotic breeds while the rest are commercial broilers and layers (Magothe et al., 2011; MoLD, 2006). The gaining popularity of poultry production in developing countries is due to its roles in bridging the protein malnutrition and economic empowerment of resource poor segment of the society (King'ori, 2011). However, there is low productivity of chicken in Kenya and other parts of the world which is partly attributed to poor management practices, such as, lack of proper health care, poor nutrition, and housing (Magothe et al., 2011). Birds primarily depend on scavenging feed resources, which mostly are inadequate in nutrient supply and highly variable. Improving poultry management particularly by targeting nutrition through supplementation can guarantee increased productivity (King'ori et al., 2010; Ndegwa et al., 1996). Excessive use of antibiotics to treat bacterial infections in poultry may lead to drug resistant bacterial strains reaching human beings. Hence feeding insects to chicken may result to decreased use of antibiotics in poultry production due to improved immunity.

2.3 Chicken Production Systems in Kenya

Production objectives, flock management, input and output levels can be used to classify chicken production systems in Kenya. As a result, we have commercial system, free range system, intensive system, and semi-intensive system. All these production systems are most common in rural areas while most farmers in urban areas practice intensive and semi-intensive system only while few practice commercial system and free-range system. The main aim of keeping chicken include subsistence or family use only, subsistence and cultural use, home consumption and income, and income only (Magothe et al., 2012b). Rearing chicken in free range system (FRS) has more returns as compared to semi-intensive system (SIS) and intensive system (IS). However, due to reduced land size, IS is commonly practiced as compared to FRS because of high human population thus leading to reduced land size (Magothe et al., 2012b). In IS, one can keep between 5 and 500 birds which are completely confined in constructed houses and offered commercial or home-made feed rations and health care. As a result, birds are protected from thieves and predators, birds are mostly kept for sale. In this system, birds show high growth rate, high egg production and low mortalities. However, it is not common in rural areas but widely practiced in urban and peri-urban areas because of high inputs required and high level of management (Magothe et al., 2012b).

Semi-intensive system requires low to medium inputs depending on the value attached to the birds. Small flocks of chicken (5 to 50 birds) are kept mainly for family use and sale. Birds are allowed to move around the homestead or in fenced runs to feed on grass, insects, kitchen wastes and any other available feed resource (King'ori et al., 2007; Magothe et al., 2012b; Mwamachi et al., 2000). However, they are given housing which range from simple shelters to proper chicken houses. The birds' health care depends on the commercial value attached to the business, but water and additional feeds are provided. As the level of input is low, production level is lower than in IS. It is a common system in highly human populated rural and peri-urban areas (Magothe et al., 2012a). Free range system is commonly practiced in low human population density rural areas and management is on low input-low output basis. Birds are kept mainly for eggs and meat for family intake, small income source and various socio-cultural uses. Mostly, small numbers of less than 30 adult birds per household are kept with less care and minimum feed supplementation (Magothe et al., 2012b; Ndegwa et al., 1998; Nzioka, 2000). In this system, feed resources mainly include grass, insects, left-over food and various seeds. Due to low inputs, production is also low but the cost per unit of egg or meat is nearly negligible (Magothe et al., 2012b; Okitoi et al., 2000).

2.4 Poultry Nutrition and Feeding

Domestic chicken (*Gallus gallus domesticus*) have been kept in Kenya for many decades for the following reasons; social-cultural, nutritional and economic uses. Despite the increasing demand for chicken products (meat and eggs) by local consumers, there is low productivity due to high disease incidences, inadequate nutrition, low genetic ability, and poor marketing channels thus reduced contribution to rural development (Magothe et al., 2012b). Commercial chicken, pheasants, turkeys, and other related poultry nutritional requirements have already been estimated (Blum et al., 1975; Potter & Shelton, 1979; Woodard et al., 1977; Yamane et al., 1980; NRC, 1994). Protein and energy requirements information of indigenous and exotic chicken is however still limited. Knowledge of protein requirements of chicken will allow producers to strategically offer supplementary sources of protein to meet any deficits. The additional supplements are meant to increase productivity of most chicken under scavenging conditions (King'ori et al., 2003, 2010).

Proper hens' nutrition is a pre-requisite for high egg production and experts have designed feeds to meet laying hens' requirements of energy, protein, mineral and vitamin depending on maintenance, body weight and egg production level of hens. Calcium, choline, vitamin A and vitamin D are specific requirements which have been demonstrated to be essential for hens (Leeson, 2010). In many developing countries, the most commonly used protein-rich feed ingredients are plant-based, including oil cake, leguminous seeds such as soya beans, and grain such as corn. However, use of soybean meal has become expensive due to the high cost of this ingredient, being sold at Kes 38,900 per metric tonne (Secci et al., 2018). Price of feed has a great influence on egg production costs, totaling to 55% to 60% in cage housing systems (International Eggs Commission (IEC), 2015; Secci et al., 2018). Prolonged feed resources shortage as a result of land scarcity due to high population growth and climate change could also have great implications for feed and food production in developing countries. Finding alternative sustainable feed options is therefore an urgent priority (Secci et al., 2018). Table 2.1 outlines the nutrient requirements of growing pullets which can replace old laying hens in commercial layers production.

Table 2.1: Nutrient requirements of growing pullets

Nutrients	Chick starter 1	Chick starter 2	Grower	Developer	Pre-lay
ME kcal/kg	2,810 – 2,920	2,810 – 2,920	2,790 – 2,900	2,710 – 2,820	2,735 – 2,930
CP %	20.0	18.3	17.5	16.0	16.5
Methionine %	0.5	0.5	0.4	0.3	0.4
Total sulfur amino acids %	0.9	0.8	0.7	0.7	0.7
Lysine %	0.9	0.9	0.8	0.7	0.7
Ca. (%)	1.0	1.0	1.0	1.4	2.5
Available P. %	0.4	0.4	0.4	0.5	0.5
Linoleic acid %	1.0	1.0	1.0	1.0	1.0

Source: NRC (1994)

2.5 Factors Affecting Chicken Performance

Performance of chicken is dependent on the following factors: growth performance of the flock, average daily gain, feed consumed and feed conversion efficiency. However, there are various problems affecting chicken farming today which may lead to great losses to farmers. According to Mapiye and Sibanda (2005), the major constraints to village chicken farming include predation, diseases and high chick mortality rate. As a result, farmers are advised to take extra care of their chicks using locally made hay box brooders which will in turn lead to reduced chick mortality. Diseases are the most important causes of economic losses in traditional poultry production systems where vaccination occurs only in response to an outbreak. Therefore, there is need to determine the efficacy and veterinary properties of ethno-veterinary medicine in village production (Weyuma et al., 2015). Other constraints may include, inadequate veterinary services, shortage of feed, inadequate resources to be used in chicken farming and lack of enough knowledge on chicken production (Weyuma et al., 2015).

2.6 Protein Feed Ingredients

Plant and animal origin feed ingredients are used as protein sources when formulating poultry feeds. Plant protein ingredients include the following: sunflower meal, soybean meal, canola meal, beans and peas, while meat meal and FM are the common animal protein sources (Ravindran, 2013). Approximately 90% of produced soybean is used in poultry diets while the rest is utilized in other livestock feed and human food (Onsongo et al., 2018). Animal origin protein sources have traditionally been used for animal feeding. However, they pose a high risk of introducing diseases to animals through this practice. Consequently, feed ingredients containing or contaminated by animal matter from any source must not be fed to livestock in Africa. In addition, many of the traditional ingredients used in poultry diets are foreseen to be in short supply within ten years. Thus, there is need to identify alternative protein sources that meet dietary requirements and reduce feed costs (Al-Qazzaz et al., 2016).

2.6.1 Fishmeal (*Rastrineobola argentea*)

Fishmeal is the best animal protein source with excellent nutritive properties that are indispensable, particularly for use in fish and poultry feeds. These advantages, together with the current laws forbidding the use of most meat meals due to problems of food security and zoonotic diseases, make it the most used animal protein source as livestock feed. The most common ingredients like FM, fish oil, soybean meal and grains use is on the rise in both human food and animal feed (Barragan-Fonseca et al., 2017; Van Huis, 2013). Fishmeal is a resource that depends on the catch and as a result it is variable quantitatively and qualitatively in terms of production (Barragan-Fonseca et al., 2017). In addition, deterioration of the marine environment and stripping of fisheries have resulted in decreased FM production and an increase in the price from Kes 60,000/MT in 2005 to Kes 200,000/MT in June 2010 (Manzano-Agugliaro et al., 2012). This increase in prices is likely to continue and with consequent economic repercussions on animal production. This situation as a result reveals the importance of renewable sources of proteins, which are particularly important in the diet of fish and poultry (Manzano-Agugliaro et al., 2012). The table below highlights the nutrient composition of FM as documented by NRC (1994).

Table 2.2 Nutrient composition of fishmeal (*Rastrineobola argentea*)

Nutrient	Composition (%)
ME (kcal/kg)	2720.0
CP	60.0
EE	2.0
Ca	6.5
Available P.	3.5
Linoleic acid	0.3
Lysine	5.3
Methionine	1.8
Threonine	2.9
Tryptophan	0.6

Source: NRC (1994)

Fishmeal prices are on the rise for instance, increased fish demand in 2010 and 2011 led to sharp higher price and although demand softened in late 2011 and early 2012, prices remained high (FAO, 2012). For small scale farmers, this means that FM will be less accessible and at the same time, aquaculture is the fastest growing animal feed producing sector and will need to expand sustainably to keep up with the increasing demand for fish.

2.6.2 Soybean (*Glycine max*)

Soybean and its' by-products demand in Kenya has risen hence exceeding local production, global land availability for soy cultivation is also limited and overfishing in marine has led to a reduction of the most common small pelagic forage fish used for fishmeal and fish oil production (Veldkamp et al., 2012). Soybean is processed either at industrial or non-industrial level with almost all of it undergoing the latter (Chianu et al., 2014). The main consumer of soybean and its by-products is the livestock feed industry (Chianu et al., 2014). Soybean meal (SBM) has relatively high CP level, good amino acid profile and high digestibility making it standard as compared to other plant protein sources. The following factors influence the nutritive value of SBM; climate, agronomic practices, cultivar, soil conditions, extent of dehulling and processing conditions (Onsongo et al., 2018). Proximate composition and mineral levels of SBM are affected by date of planting, rate of seeding and planting row type (Bellaloui et al., 2015). Significant different CP levels have been observed in SBM from USA (47.3%) and Argentina (46.9%) (Onsongo et al., 2018). Antinutritive factors in SBM include; lectins, protease trypsin inhibitors, isoflavones and oligosaccharides. Protease trypsin inhibitors are mostly considered during evaluation of SBM nutritive value (Onsongo et al., 2018).

2.7 Non-Conventional Animal Feeds for Chicken

Non-conventional animal protein (NCAP) sources such as insects (locusts, grasshopper, BSFL, termites and crickets) are principally behind the drive of improving nutritional status of chicken and improved flock productivity (Ncobela & Chimonyo, 2015). There is need to ensure sustainable production of NCAP sources in both small and large scale considering that most of the insects are seasonal. Rearing, harvesting, processing and storing methods are needed to ensure easy accessibility and harvesting of insects in large quantities, all seasons and store them safely for future use. Locally available facilities should be used for rearing, harvesting, processing and storing NCAP while observing good hygiene.

The use of NCAP is imperative as an effort to counter balance the high feed cost that account for between 60 and 70% of total cost of chicken production. The outputs generated in smallholder chicken production systems are remarkably lower than input costs, hence lower profitability (Ncobela & Chimonyo, 2015). Since most of the NCAP sources are locally available, the costs of transporting, storing and processing could be lower (Ncobela & Chimonyo, 2015). The increased interest in understanding the contribution of NCAP sources for village chicken is motivated by the desire to produce organic chicken products such as meat and eggs (Mtileni et al., 2013). These products can fetch premium prices and enhance household income and rural livelihoods. Supply of such products in the market is however erratic, low and unreliable. The contribution of NCAP sources to the diets of scavenging chicken should therefore, be estimated and documented for future use (Ncobela & Chimonyo, 2015).

2.7.1 Insects as Food and Feed

Global human population is expected to exceed 9 billion by 2050, and so the need for food, fuel, fibre and shelter will need to be met with minimal ecological footprint (Anankware et al., 2015). Feeding this rapidly growing population has implications for how we deal with food security issue now and in future. Insects have been used as food source for humanity since the first bipedal human ancestor came across the Savannah (Anankware et al., 2015). Nutrient composition analysis of commercially bred insects suggests that, they are excellent sources of most nutrients including amino acids, minerals, vitamins and fatty acids (Finke, 2015).

Calcium, vitamins D, A, E, thiamin and omega-3 fatty acids are nutrients which appear to be low across most commercially bred insect species. Other nutrient concentrations like pyridoxine and some minerals are low in certain species of commercially bred insects and this is supported by reports of proven or suspected nutrient deficiencies in captive bred insects including calcium, vitamin A, vitamin D and thiamin (Finke, 2015).

Conventional feed resources such as FM and soybean meal costs are very high and their availability in future is expected to be limited, as a result, insect rearing could be part of the solution. Studies on evaluation of insect larvae or insect meal as an ingredient in the diet of animals are limited. Nutritional composition of black soldier fly larvae, house fly maggots, mealworm, locusts, grasshoppers, crickets, and silkworm meal and their use as a replacement of FM and/or soybean meal in the diets of fish, poultry, pigs and ruminants have been discussed by Finke (2015). The CP and lipid contents of these insects are high that is, 42 to 63% and up to 36% oil respectively. The concentration of unsaturated fatty acid is high in housefly, maggot meal, worm meal and house cricket (60-70%), while black soldier fly larvae have a lower concentration of 19-37%. Several studies have confirmed that palatability of these insects larvae meal to most animals is good and they can replace 25 to 100% of soybean meal or FM depending on animal species. However, other insect meals except silkworm meal are deficient in lysine and methionine thus their supplementation in animal feed can enhance performance of different animals. Calcium is also deficient in most insect meals and its supplementation in the diet is important, especially for growing animals and laying hens. Enhancing Ca and fatty acids levels of insect meals can be achieved by manipulating the substrate used during insects rearing (Makkar et al., 2014). Table 2.3 shows the documented nutrient composition of different insect meals.

Table 2.3 Nutrient composition of different insect meals

Insect/Item	Field cricket		Housefly		BSFL	Cockroach	Mealworm
	Imago	Sub-imago	Pupae	Larvae	Larvae	Nymph	Larvae
Per kg DM							
Gross energy(MJ)	21.5	19.3	20.1	20-24	22.1	-	26.8-27.3
CF (g)	70.0	94.0	157.0	16-86.0	70.0	86-89.0	51-88.0
Ash (g)	64.0	54.0	55-98.0	31-173.0	146-284.0	46-54.0	10-45.0
P (g)	8.0	8.6	-	9.2-24.0	6.4-15.0	0.6-0.7	4.4-14.2
Ca (g)	9.9	3.1	-	3.1-8.0	50-86	0.2	0.3-6.2
CP (g)	564.0	638.0	630-762	380-604	411-450	543-734	451-603
Crude fat (g)	238	168	144-161	90-260	150-350	176-261	250-431
Glycine (g)	3.0	2.7	3.9-4.3	3.7-5.1	5.1	4.6-4.8	3.9-5.6
Arginine (g)	3.7	3.3	4.2-5.9	3.7-5.8	4.8-8.0	3.8-5.6	3.8-5.6
Threonine (g)	2.1	1.9	3.0-3.4	2.0-4.4	1.3-4.8	2.5-3.3	3.5-4.4
Valine (g)	3.4	3.0	3.4-4.6	1.3-5.1	5.6-9.1	4.4-5.1	5.5-6.6
Methionine (g)	0.8	0.8	1.5-2.6	1.3-4.6	1.4-2.4	1.1-1.2	1.1-2.0
Cysteine (g)	-	-	0.4	0.5-1.0	0.1	-	0.8-0.9
Leucine (g)	4.2	3.6	4.9-5.4	4.5-7.8	6.6-8.4	4.7-5.8	6.7-10.6
Lysine (g)	3.2	2.9	4.8-6.5	5.0-8.2	5.5-8.0	4.0-4.9	4.6-6.1
Total Omega 3 (g)	22.0	17.0	-	-	2.0	1.0-11.0	2.0-4.0
Total Omega 6 (g)	314.0	352.0	-	-	23.0	35.0-207.0	81.0-93.0

Source: Józefiak and Engberg (2015)

2.7.2 Insects as Chicken Feed

Insects have great potential as chicken feed taking into account their nutritional value, low space requirement and their great acceptance for use in poultry and fish diets as well as reptiles since insects belong to their diet in the natural habitat. In addition, insects used for feed can be raised on organic wastes such as manure and fish offal without any major problems and ethical issues (Mitsuhashi, 2010). Grasshoppers, crickets, cockroaches, termites, lice, stink bugs, cicadas, aphids, scale insects, psyllids, beetles, caterpillars, flies, fleas, bees, wasps and ants have all been used as complementary food sources for poultry and other livestock. Insects play an essential role in minimizing food insecurity by reducing FM use in animal feed in addition to providing ecosystem services such as pollination, waste degradation and biological control. Van Huis (2013), outlined the important role of insects in assuring food and feed security. It is believed that 1900 species of insects are consumed by about 2 billion people globally, mainly in developing world. This is very important as the need for alternative protein sources is on the rise due to rapid urbanization in developing countries and the shift in composition of global food demand (Kelemu et al., 2015; Van Huis, 2013).

Increasing demand for alternative protein sources world-wide, has led to improved awareness for insects use to represent an innovative food and feed source rich in high quality protein as well as other beneficial nutritional ingredients such as fat, minerals and vitamins has been raised. Despite traditional knowledge about insects and their harvest in the wild, industrial mass production of safe insects and insect products for consumption and for processing into food and feed is necessary. Development of rearing, harvesting as well as post-harvest technologies for these insects is required (Birgit et al., 2013; Makkar et al., 2014).

2.7.3 Mass Rearing of Insects for Food and Feed

Mass rearing of insects can have a positive impact on the environment than conventional livestock production (Oonincx et al., 2010; Van Huis, 2013). This can be achieved by recycling organic by-products to high quality compost fertilisers which can be used in farming. Insects have been shown to play such an important role as well as utilisation of maggots or larvae directly as animal feed (Čičková et al., 2012; Kelemu et al., 2015). Food security and livelihood improvement of the rural poor can be achieved by practising semi-cultivation and harvesting of edible insects which have the possibility of contributing to habitat conservation. During insect rearing, women participate in larvae and pupae collection, which has been reported to provide cash income for basic expenditure, education, food and purchasing farm inputs (Agea et al., 2008; Hope et al., 2009; Kelemu et al., 2015). With the growing agribusiness companies in Africa where insects are used as food, feed and in waste conversion (Agbidye & Nongo, 2009; Agea et al., 2008; Ayieko & Nyambuga, 2009; Kelemu et al., 2015), insects can play a clear role in eradication of food insecurity, hunger and malnutrition in Africa (Ayieko et al., 2010; Kelemu et al., 2015;Vantomme et al., 2004).

While most insects are commonly harvested in the wild, bees and silkworms, have been domesticated for a long period of time because of the value of their products. Other insects are raised in large numbers for the purposes of pollination, biological control such as predators, parasitoids and health (maggot therapy). Keeping insects for food is a relatively new concept and cricket farming in the Lao People's Democratic Republic, Thailand and Vietnam is an example of insect reared for human consumption in the tropics. In temperate zones, insect rearing is carried out chiefly by family run enterprises who keep insects such as grasshoppers, crickets and mealworms in large quantities as pets or for zoos. Commercialisation of insects as feed and food by commercial firms only took place recently and research on insects intended for human consumption is still minimal (Van Huis et al., 2013).

2.7.4 Challenges of Insect Rearing, Processing and Storage

Insect meal can fully replace either FM or soybean meal in poultry feed formulation when large insect quantities have been reared at a low cost (Onsongo et al., 2018; Van Huis et al., 2013). However, successful rearing of insects requires critical element such as research on insect biology, control of rearing condition and diet formulae for the farmed insect species. Most of the available insect production systems are expensive thus posing a challenge for mass insect rearing. Another major challenge of such industrial scale rearing of insects is the development of automation processes which makes plants economically competitive with the production of meat (meat-substitutes like soy) from traditional livestock or farming sources (Van Huis et al., 2013).

Insect rearing is expensive due to high necessity for manual labour during mass production. Also, facilities and resources required for mass insect rearing have not been put into consideration. However, technological improvement of insect rearing facilities, cheaper production processes, hygienic measures and sanitary standards are key for disease and contamination prevention during mass insect rearing (Rumpold & Schluter, 2013).

2.8 Black Soldier Fly (BSF)

Black soldier fly (*Hermetia illucens*) is a fly of the Stratiomyidae family and are found in abundance (where they naturally occur) around poultry, pigs and cattle manure piles, hence they are known as latrine larvae (Anankware et al., 2015; Van Huis et al., 2013). It is a tropical fly which was first seen in 1930 in Islands. It is said to have moved from Argentina to Boston and Seattle and later spread to Europe, India, Asia and Australia during World War II. Black soldier fly larvae can occur in very dense population of organic wastes such as coffee bean pulp, vegetables, distillers' waste and fish offal (fish processing by-products). It is mostly considered a good feed protein ingredient because of its ability like other insect species to convert large amounts of organic waste (about 1.3 billion tones every year) into protein rich biomass with excellent amino acid profile (Maurer et al., 2015; Onsongo et al., 2018). These insects are often found around decaying organic matter for example animal manure or plant material.

They consume decaying matter thus have shown to reduce animal waste in commercial swine and poultry facilities into organic fertilizer and animal protein (Diclaro & Kaufman, 2009; Newton, 2005). These insects are able to produce antimicrobial peptides (Ratcliffe et al., 2014) used to protect them from contaminations and also reduce harmful bacteria in manure (Erikson et al., 2004; Marono et al., 2017) additionally, during feed production, different systems of decontamination should be used by producers to reduce losses (Rumpold & Schluter, 2013). These insects can be used commercially to solve environmental problems which include manure and other organic wastes such as, manure mass reduction, moisture content minimization and offensive odour reduction. They also provide high-value feedstuff for cattle, pig, poultry and fish. The adult black soldier fly is not common in human habitats or foods thus they are not considered a nuisance to the environment. They also contain high crude fat content which can be converted to biodiesel: for instance, 1000 larvae grown on 1kg of cattle manure, pig manure and chicken manure can result to 36g, 58g and 91g, respectively, of biodiesel (Li et al., 2011). Black soldier fly larvae meal also contains chitin, a polysaccharide present in arthropod's exoskeleton. Chitin is not digestible by monogastric animals (Sanchez-Muros et al., 2014) and can negatively affect the protein digestibility (Longvah et al., 2011) but may have a positive effect on the functioning of poultry immune system (Bovera et al., 2016; Sanchez-Muros et al., 2014). Insects have a higher feed conversion efficiency compared to conventional livestock, (Nakagaki & Defoliart, 1991; Veldkamp et al., 2012) and their fecundity is very high. Studies have shown that insects might produce less greenhouse gases as compared to pigs and cattle (Oonincx et al., 2010).

2.8.1 Life Cycle of Black Soldier Fly

Some BSF have different colors which range from yellow, green, black, blue and some have a metallic appearance. Other members mimic other insects such as bees and wasps. Physiology of BSFL has been widely described by Gujarathi and Pejaver (2013). Adult flies do not possess bristles and have wasp like appearance. In most cases, adult female BSF lay eggs on, in, or around wet organic material such as carrion (Bondari & Sheppard, 1987; Nyakeri et al., 2017) and common agricultural sites where huge amounts of organic material left by livestock provide ample sites for their reproductive needs (Nguyen-Viet et al., 2009; Nyakeri et al., 2017). However, in natural breeding sites they lay their eggs in moist organic material and in urbanized areas eggs are laid in dump sites or compost, which offer nutritional needs to the young hatched larvae to eat and grow (Diclaro & Kaufman, 2009).

Black soldier fly larvae generally pass through four life stages and these are egg, larvae (five instars), pupa and adult stage (Hall & Gerhardt, 2002). When mature they are about 25 mm in length, 6 mm in diameter and weigh approximately 0.2g. These larvae and pupae are very tough, robust and can survive under conditions of extreme oxygen deprivation. The larvae are pale white in colour and have a small black head containing their mouthparts (Newton et al., 2005).

2.8.2 Black Soldier Fly Entomology and Distribution

It is believed that BSF originates from South America and can be commonly found in warm temperate climates (Žáková & Borkovcová, 2013). However, this insect has been reported to be available in almost 80% of the world Africa included (Olivier, 2009). In Africa, BSF is common in Ghana and South Africa as the larvae is used for compositing organic matter. However, in Kenya there is various documented evidence of the presence of these insects (Nyakeri et al., 2017). In south eastern United States, these insects are abundant during late spring and early fall while in Georgia it has three generations per year. However, they are found throughout the Western Hemisphere in the continental United States (Diclaro & Kaufman, 2009; Tomberlin et al., 2002).

2.8.3 Nutritional Attributes of BSFL

Black soldier fly larvae is a high value feed source rich in protein and fat, it contain about 40-50% CP. The amount of fat is extremely variable and depends on the type of substrate used to rear BSFL. For instance, its fat content is reported to be 15-25% DM (larvae fed on poultry manure), 28% DM (swine manure), 35% DM (cattle manure) and 42-49% DM (oil-rich food waste) (Newton et al., 2005) as shown in table 2.4.

Table 2.4: Crude protein and crude fat content of black soldier fly larvae reared on different substrates

Substrate	%CP	%CF	References
Cattle manure	42.1	29.9-34.8	Li et al., 2011b; Newton et al., 1977
Chicken manure	40.1	27.9	Li et al., 2011b; Manzano-Agugliaro et al., 2012; Newton et al., 2005
Swine manure	43.2-43.6	26.4	St-Hilaire et al., 2007;
Palm kernel meal	42.1-45.8	27.5	Rachmawati et al., 2010
Restaurant waste	-	39.2	Zheng et al., 2012
Chicken feed	47.9	14.6	Bosch et al., 2014; Nguyen et al., 2015; Oonincx et al., 2015
By products[1]	41.9	-	Oonincx et al., 2015
Liver	62.7	25.1	Nguyen et al., 2015
Fruits and vegetables	38.5	6.63	Nguyen et al., 2015
Fish	57.9	34.6	Nguyen et al., 2015

[1]Beet molasses, potato steam peelings, spent grains and beer yeast, bread and cookie remains.
Source: Barragan-Fonseca et al. (2017)

The ash content of these insects is relatively high but variable, from 11 to 28% DM. Also, larvae is rich in calcium (5-8% DM) and phosphorus (0.6-1.5% DM) (Gutierrez et al., 2004; Newton et al., 1977; St-Hilaire et al., 2007; Yu et al., 2009) and lysine (6-8% of the protein). The dry matter content of fresh larvae is high, that is 35-45%, which makes them easier and less costly to dehydrate than other fresh by-products. Black soldier fly larvae tend to contain less protein and more lipids than housefly maggots (*Musca domestica*). Fatty acid composition of the larvae depends on fatty acid composition of the diet or substrate used. Larvae fed cow manure contain 21% of lauric acid, 16% of palmitic acid, 32% of oleic acid and 0.2% of omega-3 fatty acids. However, for larvae fed 50% fish offal and 50% cow manure the proportions were 43%, 11%, 12% and 3%, respectively. Total lipid content also increased from 21% to 30% DM. To enrich the final biomass of black soldier fly larvae, they can be fed on a diet made of wastes containing desirable amounts of omega-3 fatty acids (St-Hilaire et al., 2007; Yu et al., 2009).

Elwert et al. (2010) study compared *Hermetia illucens* meal to fishmeal and the results showed higher levels of threonine, valine, isoleucine, leucine and histidine in the insect meal comparative to lysine but methionine level was slightly lower (Al-Qazzaz et al., 2016). Black soldier fly larvae amino acids composition and fatty acids (FA) level depends strongly on the diet fed. St-Hilaire et al. (2007) documented that BSFL fed on cow manure had a different total fat content and a different FA profile than those fed with a mixture of cow manure and fish offal. Insects fed on fish offal had higher lipid content but manipulation is required to include desirable FA, likes a-linolenic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). The omega-3 FA of the larvae are enriched and a higher total FA content is achieved (Al-Qazzaz et al., 2016). Omega-3 FA which are long chain polyunsaturated fats, have various benefits for human health, this includes, reduction of coronary heart disease risk (Kralovec et al., 2012). They are precursors to anti-inflammatory mediators, preventing inflammatory mediated disorders such as allergy, diabetes, Alzheimer's disease and Neuro-degenerative diseases (Al-Qazzaz et al., 2016).

According to Al-Qazzaz et al. (2016) and Diener et al. (2009) BSF prepupae has a chitin content of 8.72% DM. Chitin is a widely occurring polysaccharide in nature which is found in the cuticle of the BSF. It is typically found in a complex matrix with other compounds (Finke, 2007; 2015). Chitin is composed of b (1®4) linked 2-deoxy-2-acetamido-D-glucopyranosyl residues. Studies have shown, to increase the digestibility of BSFL meal, chitin can be removed by multiple mechanisms such as, alkali extraction (Al-Qazzaz et al., 2016) adding chitosan, chitooligosaccharides or acetylglucosamine (Mao et al., 2012). Addition of chitin degrading enzymes or bacteria can also improve the digestibility of chitin-protein complexes (Kroeckel et al., 2012) thus fast digestion of the meal.

2.8.4 Black Soldier Fly Larvae as Livestock Feed

Livestock production sector is faced with the problem to improve advanced methods necessary to achieve future social, environmental and economic needs. Rearing insects on organic matter can lead to appropriate alternative animal protein source for animals (Kim et al., 2011; Smetana et al., 2016). Black soldier fly larvae meal has many key advantages as compared to other insect species, when used as animal feed. It is polyphagous and contains high levels of amylase, lipase and protease activities in its gut extracts (Kim et al., 2011). As a result, it is used in animal waste recycling (Barragan-Fonseca et al., 2017; Myers et al., 2008; Newton et al., 2005; Nguyen et al., 2013), faeces (Diener et al., 2009; Lalander et al., 2013; Oonincx et al., 2015), and organic wastes (Barragan-Fonseca et al., 2017; Diener et al., 2011; Gujarathi & Pejaver, 2013; Green & Popa, 2012; Kalová & Borkovcová, 2013; Nguyen et al., 2013), converting bio-waste into high quality nutrient for use in animal feed (Barragan-Fonseca et al., 2017; Veldkamp et al., 2012).

Different studies have shown the use of BSFL as feed for different animals such as pigs, poultry, fish, crustaceans and other species. According to Newton et al. (2005) BSFL is a suitable ingredient in growing pigs' diets, because of its amino acid, lipid and calcium contents. However, additional refinement (cuticle removal and rendering) may be necessary to make prepupae BSFL suitable for chicks and early weaned pigs. Several experiments have also shown that BSFL could be a partial or full substitute for FM in fish diets (Newton et al., 2005). Other additional trials as well as economic analyses are still necessary as reduced performance has been observed in some cases. The type of rearing substrate and processing methods affect the utilization of BSF larvae by fish (Newton et al., 2005).

2.8.5 Use of BSFL in Chicken Feeds

Chicken production industry has grown rapidly in developing countries in the last two decades. Amino acids (lysine, methionine and cysteine) in poultry feed is supplied by animal and plant proteins. Animal-based, protein-rich feed ingredients generally contain imported fish and meat or blood meal, while plant-based ingredients include imported oil cakes and leguminous grains (Newton et al., 2005). However, feeding insects to chicken leads to reduced use of antibiotics in the poultry industry due to improved immunity hence less disease incidences. Excessive use of antibiotics in poultry can lead to human infection with drug-resistant bacterial strains through the consumption of chicken products (Newton et al., 2005).

Black soldier fly larvae provide high value feedstuff for poultry which can be used in feed formulation to substitute FM at different inclusion levels (Newton et al., 2005). Dried BSFL contain 42- 50% CP and 35% (on dry matter basis). Live prepupae consist of 44% dry matter and can easily be stored for long period once dry. They have been found to support good growth of growers, broilers and layer hens when used as component of a complete diet (Newton et al., 2005). However, the main hurdles for insects use in animal feed include limited quantity of produced insects, high cost for their meals and, in Europe, the Regulation EC No. 1069/2009 which forbids the feeding of farmed animals with processed animal protein (Józefiak et al., 2016). Some insects are considered more important in poultry nutrition due to their chemical-nutritional characteristics, but also its important to consider the technique used in larvae meal production (Loponte et al., 2017).

Black soldier fly larvae process organic waste fast (depending on the volume of waste offered), reduce bacterial growth and minimize production of bad odor in livestock houses.

Moreover, BSFL aerates and dries manure thus reducing odor (Van Huis et al., 2013). They are a competitor to housefly (*Musca domestica*) larvae, which make manure more liquid and thus less suitable for BSF larvae. Their presence is also believed to inhibit oviposition by housefly. For instance, they have been shown to reduce housefly population in pig and poultry manure by 94-100%. As a result, they can help to control housefly population in livestock farms and in households with poor sanitation. This will improve health status of the animals and people since housefly is a major disease vector (Newton et al., 2005; Sheppard et al., 1994).

2.8.6 Rearing of BSFL for Feed

Black soldier fly is inactive during winter months in temperate countries, thus requires rearing conducted indoors under artificial conditions (Zhang et al., 2010). In laboratory-controlled conditions BSF larvae (having six larval instars including prepupae) reach the pre-pupa stage after two weeks at 30°C. They then leave food substrate to pupate in a dry dark place (Barragan-Fonseca et al., 2017). Adult flies emerge after 10-14 days at 27-30°C (Barragan-Fonseca et al., 2017; Sheppard et al., 2002). They do not feed to survive but their life span is increased when sprayed with water mixed with sugar or honey (Barragan-Fonseca et al., 2017; Tomberlin et al., 2002). Two-day-old adults mate then females oviposit in crevices near food sources two days later (Barragan-Fonseca et al., 2017; Tomberlin & Sheppard, 2002). Egg-hatching takes place after four days at 27°C (Barragan-Fonseca et al., 2017).

The BSF life history characteristics includes, survival rate and development time. They are determined by a number of factors such as temperature, relative humidity, food availability, and food composition (Gobbi et al., 2013). They have a wide geographic distribution which shows their tolerance for a wide range of abiotic conditions. However, there are optimal values for these conditions for maximum BSF performance as shown in table 2.5.

Table 2.5: Abiotic factors for rearing black soldier fly

Factors	Min.	Optimal	Max.	References
Temperature (°C)	12	26-27	36	Booth and Sheppard, 1984; Holmes, 2010; Holmes et al., 2016; Sheppard et al., 1994; Tomberlin et al., 2009
Relative humidity (%)	25	60-70	99	Gobbi, 2012; Holmes et al., 2012; Tomberlin & Sheppard, 2002
Substrate moisture (%)	40	52-70	70	Fatchurochim et al., 1989; Furman et al., 1959; Tomberlin et al., 2002
Light intensity[1]	60	135-200	-	Alvarez, 2012; Holmes, 2010; Tomberlin & Sheppard, 2002; Zhang et al., 2010

[1]Light intensity is expressed in µmol/m2/s and affects mating and egg fertilisation of BSF. However, it has been suggested that light spectral composition plays a more important role in fertilisation than light intensity. Light-emitting diodes producing wavelengths in the UV, blue and green ranges have proved to increase the proportion of fertilised eggs (Oonincx et al., 2016).
Source: Barragan-Fonseca et al. (2017)

Black soldier fly are mostly harvested at the pre-pupae or larvae stage where they are 44% in terms of dry matter, 42% protein and 35% fat, including high fatty acids and all essential amino. Several studies have shown that BSFL is a good nutrient source for cockerels, swine, and tilapia (Bullock et al., 2013). A study conducted on channel catfish showed that replacing *Menhaden* with BSF larvae do not result in loss of growth. However, the price of FM is constantly on the rise each day, for instance, *Menhaden*, containing 65% protein is valued at about Kes 160 per kg in major Kenyan commodity markets. Black soldier fly larvae projected value is Kes 105 per kg (Bullock et al., 2013), this can significantly decrease the cost of feeds, which total to 40-70% of overall production cost. Studies indicate that over 60 tons (55 MT) of BSF prepupae could be self-harvested from a 100,000 hen caged layer house in one summer if that manure is utilised by female black soldier for oviposition (Bullock et al., 2013; Sheppard et al., 1994). However, in swine manure management trial, it was found that oviposition almost doubled the biomass observed while using layers manure (Bullock et al., 2013).

Information on BSFL rearing is very limited and it is reported that, rearing wild collected eggs to adults will take 38 days at 29.3^{0}C. Mating and oviposition can often be observed in a 3 by 6.1 by 1.8 meter cage kept outdoors. In addition, when females are held in a greenhouse, few fertilized eggs will be collected as compared to what is observed when they are kept in a 0.76 by 1.14 by 1.37 meter cage held outdoors, hence, direct sunlight is reported to boost mating. When BSFL are kept in smaller cages (53 by 91 by 53 cm and 38 by 46 by 38 cm) no mating or egg collection can be observed (Diclaro & Kaufman, 2009). In some cases, a female can lay 500 or more eggs in a dry environment near edges or crevices of decaying organic matter. These eggs are approximately 1 mm in length and creamy white in colour (Diclaro & Kaufman, 2009). After hatching, larvae consume whatever waste available for growth and survival for three weeks when they will have reached full maturity as long as environmental conditions are favourable. However, in hostile circumstances it may take up to six months for larvae to reach maturity because BSFL has the ability to extend its life cycle. Larvae can be approximately 27 mm in length, 6 mm in width and have a pale white colour with a small black head containing mouth parts (Diclaro & Kaufman, 2009; Newton et al., 2005). They are then harvested, dried, defatted and ground to be used as protein sources in animal feeds during feed mixing.

2.9 Use of BSF in Waste Management and Other Purposes

These insects are perfect converters of feedstuff or organic waste into appreciated biomass or manure. Black soldier fly has the ability to extend its life cycle when living in unfavorable environment hence it can be used in waste disposal processing (Newton et al., 2005). These insects can directly be fed to fish or can be dried and supplemented in the diet as protein source for animals. Thus, rearing BSF can help in discouraging inhabitation by other common fly species and also reduce the likelihood of disease transmission (Newton et al., 2005). Black soldier fly larvae has been used to solve some problems related to organic waste, which include offensive odour, decreasing manure mass and moisture content of organic waste (Al-Qazzaz et al., 2016; Barry, 2004; Newton et al., 2005). However, it is documented that poultry and pig manure colonization by BSFL can result to reduced population (94 to 100%) of common housefly (*Musca domestica*).

Black soldier fly larvae could be used to solve problems associated with large manure accumulation at confined animal feeding operations (Maurer et al., 2016). They have a short life cycle which encourages large scale and long-term production along with the assurance of a reliable source of food due to frequent reproduction. Generally, insects can feed on a number of materials such as manure and they are better feed convertors than most other animals, which results in reduced production cost and wastages (Khusro et al., 2012; Maurer et al., 2016). Black soldier fly has shown to be particularly promising, as it can reach high growth rate by feeding on materials that are unfit for human consumption such as, by-products from food processing and organic waste (Diener et al., 2011; Maurer et al., 2016).

Black soldier fly is an interesting insect for the conversion of organic waste. It is shown that, it can efficiently transform swine, dairy and poultry manure to body mass, reducing dry matter mass up to 58% and associated nutrients including P and N by 61–70% and 30–50%, respectively. House fly population are also decreased in chicken manure by these insects. It is also shown that BSF larvae can efficiently reduce and recycle fish offal from processing plants. It has also been reported that insects release less greenhouse gases and ammonia as compared to cattle or pigs when retained together. They also pose less risk of transmitting zoonotic diseases to wildlife, humans and livestock compared with birds and mammals (Van Huis, 2013). The larval stage of BSF is described to diminish the quantity of pathogens such as *Escherichia coli* and *Salmonella enteric* serotype enteritidis on organic compost (Bullock et al., 2013; Erickson et al., 2004; Liu et al., 2008; Nyakeri et al., 2017), hence making the residues safe for use in crop farming as organic fertiliser (Banks et al., 2014; Choi et al., 2009; Nyakeri et al., 2017).

Research Gap;

(i) To investigate BSFLM as a potential prebiotic in exotic layer diets, since chitin degradation can be associated with high production of short chain fatty acids.

(ii) There is need to establish cost effective well optimized facilities for mass rearing of BSFL to achieve high production.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

This study was conducted at Kenya Agricultural and Livestock Research Organization (KALRO) in Naivasha town, Nakuru County. The area has an altitude of 1,800 meters above sea level, 0° 43′ 12.85″ South latitude, 36° 25′ 42.71″ East longitude with mean annual rainfall of 1,000mm and 17 to 22^{o}C temperature range (Google map downloaded on 11.06.2018).

Figure 3.1: Map of Kenya showing location of KALRO Naivasha

Source: Google map

3.2 Experimental Diets

Five experimental diets containing 2800 Kcal/kg energy and CP percentage dry matter of 20%, 18% and 16% for chicks, growers and layers respectively were formulated using BSFLM to replace FM at 0%, 25%, 50%, 75% and 100% substitution levels. The experimental diets included: T1-control, (100% FM); T2, (75% FM and 25% BSFLM); T3, (50% FM and 50% BSFLM); T4, (25% FM and 75% BSFLM) and T5, (100% BSFLM), together with other feed ingredients to make a complete diet. National Research Council (NRC) (1994) poultry nutrient requirements were followed and Winfeed software (version 2.8) used to ensure the final formulated feed was iso-caloric and iso-nitrogenous. Whole maize and wheat pollard were used as energy sources. Soybean meal, FM and BSFLM were used as protein sources, while minerals and vitamins were supplemented appropriately. Whole black soldier fly larvae was sourced from the International Centre of Insect Physiology and Ecology (ICIPE) in Nairobi while other feed ingredients were purchased from local suppliers. Feed ingredients were ground, weighed and mixed thoroughly to a uniform blend using a feed mixer to ensure the final feed were mixed completely. Tables 3.1, 3.2 and 3.3 presents' ingredients composition that were used to compound experimental diets for chicks, growers and layer birds.

Table 3.1: Ingredients composition (g/kg as fed) of chick mash

Ingredient	T1(control)	T2	T3	T4	T5
Whole maize	60.0	60.0	60.0	60.0	60.0
Soybean meal	21.0	21.0	21.0	21.0	21.0
Fishmeal	10.0	7.5	5.0	2.5	0
BSFLM	0.0	2.5	5.0	7.5	10
Vegetable oil	2.0	2.0	2.0	2.0	2.0
Limestone	5.0	5.0	5.0	5.0	5.0
DCP	1.5	1.5	1.5	1.5	1.5
Iodized salt (NaCl)	0.3	0.3	0.3	0.3	0.3
Layer premix[a]	0.2	0.2	0.2	0.2	0.2

[a]Super layer premix contents per 2.5 kg: Vit. (Vitamin) A: 8000000 iu, Vit. D3:2000000 iu, Vit. E: 3000 mg, Vit. K3: 2000 mg, Vit B2: 3500 mg, Pantothenic Acid: 6600 mg, Niacin: 20000 mg, Folic Acid: 550 mg, Vit. B12: 6 mg, Choline chloride: 200000 mg, Lysine: 350 mg, Methionine: 120 mg, Manganese: 63000 mg, Iron: 23000 mg, zinc: 63000 mg, Copper: 14000 mg, Cobalt: 1000 mg, Iodine: 2000 mg, Selenium: 100 mg and BHT: 120000 mg. BSFLM- Black soldier fly larvae meal, Ca- calcium, DCP- Dicalcium phosphate, FM- fishmeal, T1- 0% BSFL, T2- 25% BSFLM and 75% FM, T3- 50% BSFLM and 50 % FM, T4- 75% BSFLM and 25% FM and T5- 100% BSFLM.

Table 3.2: Ingredients composition of growers mash

Ingredient (kg)	T1(control)	T2	T3	T4	T5
Whole maize	50.0	50.0	50.0	50.0	50.0
Pollard (wheat)	19.0	19.0	19.0	19.0	19.0
Soybean meal	13.0	13.0	13.0	13.0	13.0
Fishmeal	10.0	7.5	5.0	2.5	0.0
BSFLM	0.0	2.5	5.0	7.5	10
Limestone	5.5	5.5	5.5	5.5	5.5
DCP	2.0	2.0	2.0	2.0	2.0
NaCl	0.3	0.3	0.3	0.3	0.3
Layer premix[a]	0.2	0.2	0.2	0.2	0.2

[a] Super layer premix contents per 2.5 kg: Vit. (Vitamin) A: 8000000 iu, Vit. D3:2000000 iu, Vit. E: 3000 mg, Vit. K3: 2000 mg, Vit B2: 3500 mg, Pantothenic Acid: 6600 mg, Niacin: 20000 mg, Folic Acid: 550 mg, Vit. B12: 6 mg, Choline chloride: 200000 mg, Lysine: 350 mg, Methionine: 120 mg, Manganese: 63000 mg, Iron: 23000 mg, zinc: 63000 mg, Copper: 14000 mg, Cobalt: 1000 mg, Iodine: 2000 mg, Selenium: 100 mg and BHT: 120000 mg. BSFLM- Black soldier fly larvae meal, Ca- calcium, DCP- Dicalcium phosphate, FM- fishmeal, T1- 0% BSFLM, T2- 25% BSFLM and 75% FM, T3- 50% BSFLM and 50 % FM, T4- 75% BSFLM and 25% FM and T5- 100% BSFLM.

Table 3.3: Ingredients composition of layers mash

Ingredient (kg)	T1(control)	T2	T3	T4	T5
Whole maize	53.0	53.0	53.0	53.0	53.0
Pollard (wheat)	18.0	18.0	18.0	18.0	18.0
Soybean meal	10.0	10.0	10.0	10.0	10.0
Fishmeal	10.0	7.5	5.0	2.5	0
BSFLM	0.0	2.5	5.0	7.5	10
Limestone	6.5	6.5	6.5	6.5	6.5
DCP	2.0	2.0	2.0	2.0	2.0
NaCl	0.3	0.3	0.3	0.3	0.3
Layer premix[a]	0.2	0.2	0.2	0.2	0.2

[a] Super layer premix contents per 2.5 kg: Vit. (Vitamin) A: 8000000 iu, Vit. D3:2000000 iu, Vit. E: 3000 mg, Vit. K3: 2000 mg, Vit B2: 3500 mg, Pantothenic Acid: 6600 mg, Niacin: 20000 mg, Folic Acid: 550 mg, Vit. B12: 6 mg, Choline chloride: 200000 mg, Lysine: 350 mg, Methionine: 120 mg, Manganese: 63000 mg, Iron: 23000 mg, zinc: 63000 mg, Copper: 14000 mg, Cobalt: 1000 mg, Iodine: 2000 mg, Selenium: 100 mg and BHT: 120000 mg. BSFLM- Black soldier fly larvae meal, Ca- calcium, DCP- Dicalcium phosphate, FM- fishmeal, T1- 0% BSFLM, T2- 25% BSFLM and 75% FM, T3- 50% BSFLM and 50 % FM, T4- 75% BSFLM and 25% FM and T5- 100% BSFLM.

3.3 Experimental Chicken and Design

3.3.1 Chick and Grower Experiment

Three hundred (n=300) day old Isa Brown chicks were sourced from Kenchic limited, Kenya and adapted for 7 days. During the acclimatization period, all chicks were kept together in a round deep litter wooden brooder. Wood shaving was used as litter where the brooder floor was covered with about 3 inches of the shavings. Three infra-red bulbs of 250 Watts were suspended 50cm over the brooder and used as heat source and light was provided for 24 hours. After one week, all the chicks were weighed, initial weight recorded and 250 chicks randomly moved to metallic cages (each cage having 5 birds). They were then randomly assigned to five treatment diets (T1, T2, T3, T4, T5), where birds were offered the following feed specifications; chicks 35-70g, growers 70-95g, pre-laying 95-110g for a period of 20 weeks. The feeding experiment adopted a completely randomized design (CRD) with 5 birds per cage and each treatment was replicated 9 times.

3.3.2 Layer Experiment

A total of seventy-five (n=75) Isa Brown layer hens were used which were assigned to five experimental diets, each replicated 3 times with 5 hens per replicate in a completely randomized design. Hens were offered between 110-150g of feed daily for a period of 25 weeks. Water was offered *ad libitum* and other management practices remained constant for all the hens. Table 3.4 shows the experimental layout of the layers' trial

Table 3.4 Experimental layout table

Replicates	Treatments				
	1	2	3	4	5
1	T1	T2	T5	T4	T3
2	T1	T5	T3	T2	T4
3	T3	T1	T4	T5	T2

T1-control (100% fishmeal); T2 (75% fishmeal and 25% BSFLM); T3 (50% fishmeal and 50% BSFLM); T4 (25% fishmeal and 75% BSFLM) and T5 (100% BSFLM).

3.4 Data Collection

3.4.1 Growth and Feed Intake

Chick, grower and layers mash were formulated and offered to the birds during the feeding experiment for a period of 45 weeks. Management practices such as vaccination, treatment and deworming were carried out according to Kenchic limited guide. The following measurements were taken; initial weight, average daily weight gain (measured on a weekly basis) and final weight. Overall weight gain was calculated as final weight minus initial weight. Feed conversion ratio was determined as feed intake divided by weight gain. The weight of feed consumed was calculated as feed offered minus feed refusal. All these were done for chick, grower and layer stages of the feeding experiment.

3.4.2 Nutrient Analysis of Diets and Ingredients

All the five diets and some ingredients were milled to pass through a 1mm sieve for chemical analysis. Dry Matter (DM), nitrogen (crude protein), crude fat and ash content were determined according to the standard methods of AOAC (1990). Crude protein of the feed samples was determined using Kjeldahl nitrogen method and calculated as N x 6.25. Black soldier fly larvae meal and FM samples were analysed to determine amino acid concentrations. Oxidation and hydrolysis method (AOAC Official Method 994.12 "Amino Acids in Feed", Alternatives I und III) was used for the following amino acids; methionine, cystine, lysine, threonine, arginine, isoleucine, leucine, valine, histidine, phenylalanine, tyrosine, glycine, serine, proline, alanine, aspartic acid and glutamic acid (AOAC, 1995).

3.4.3 Laying Capacity

Number of eggs laid was recorded on a daily basis. Hen day egg production (HDEP) was calculated using the following formula (NRC, 1994):

$$\text{HDEP} = \frac{\text{Number of eggs produced daily}}{\text{Number of birds in the flock that day}} \times 100$$

3.4.4 Egg Characteristics

Five eggs per treatment were randomly sampled weekly from 20 weeks onwards for measurement. Both external and internal egg characteristics were measured. Egg weight was measured using an electronic digital (SHIMADZU-TXB6201L) scale. Other external egg parameters including egg length, egg width and shell thickness were measured using a Vernier caliper. The egg was then broken open on a plate to measure internal egg qualities using a Vernier caliper. They included albumin and yolk height, which were measured as described by Haugh (1937). Also, length of egg white and yolk length were measured, and the mean diameter calculated. Yolk color was determined by comparing the color of egg yolk placed on a white plate with a Roche color chart which consisted of 1-15 strips ranging from pale to orange yellow color. Yolk index was used to determine egg freshness while shape index was for egg shape determination. Haugh unit was used for egg quality determination. They were calculated as follows:

$$\text{Yolk index } (\%) = \frac{\text{Yolk height (mm)}}{\text{Yolk diameter (mm)}} \times 100$$

$$\text{Shape index } (\%) = \frac{\text{Egg width (mm)}}{\text{Egg length (mm)}} \times 100$$

$$HU = 100^{*}\log (h - 1.7w^{0.37} + 7.6)$$

Where:

HU- Haugh unit;

h = Observed height of albumen (mm);

w = Average egg weight (g).

3.5 Economic Analysis

The cost benefit ratio (CBR) and return on investment (RoI) were used in assessing the economic effect of substituting FM used in layer hens' diets with BSFL meal. Total cost of production included medication, feed, labor, housing, water, electricity, feeders and drinkers among others. However, only the cost of feed was used to determine economic costs as the rest were assumed to be constant for all the treatments. Ingredient prices based on quantities of each added in each treatment were used to calculate feed costs. Return on investment is used to measure the gain/loss produced by an investment relative to the money invested. The higher the RoI value the better the RoI of the project under consideration (De-pach, 2012). The following formulas were used;

$$Return\ on\ investment\ (RoI) = project\ revenue - project\ cost/project\ cost\ X\ 100$$

$$Cost\ benefit\ ratio\ (CBR) = project\ revenue/\ project\ cost$$

3.6 Statistical Analysis

Data on daily weight gain, initial weight, final weight, feed intake, egg characteristics, HDEP, and FCR were analyzed using one-way analysis of variance (ANOVA) in SAS version 9.00 (2002) with a completely randomized design (CRD) model. In this study, each cage represented an experimental unit while each egg was an experimental unit for egg quality characteristics analysis. The significance between treatment means was tested at statistical significance level of 5% and separated using Tukey in case of significant difference.

The model used for this experiment was;

$\mathbf{Y_{ij}}$ **=** $\boldsymbol{\mu + \tau_i + \varepsilon_{ij}}$ where:

$\mathbf{Y_{ij}}$ = Overall observation from the **i**th treatment and **j**th replication,

$\boldsymbol{\mu}$ = overall mean,

$\boldsymbol{\tau_i}$ = the treatment effects,

$\boldsymbol{\varepsilon_{ij}}$ = random error term.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Chemical Composition of Experimental Diets

The chemical composition of chick mash, growers mash and layers experimental diets used in the study are presented in Table 4.1, 4.2 and 4.3 respectively.

Table 4.1 Chemical composition (% DM basis) of chick mash

Chick mash	T1	T2	T3	T4	T5
Energy Kcal\kg	2904	2875	2871	2845	2872
Dry matter (%)	91.6	91.7	91.5	92.4	92.2
Ash (%)	7.0	6.8	6.7	6.4	6.4
CP (%)	19.4	20.6	19.5	19.7	19.9
EE (%)	8.1	8.1	7.0	10.3	11.1
NDF (%)	27.7	32.2	34.2	31.3	34.4
ADF (%)	12.3	13.9	15.3	12.2	13.5
Calcium (%)	4.5	4.5	3.3	3.5	3.2

Table 4.2 Chemical composition (% DM basis) of growers mash

Growers mash	T1	T2	T3	T4	T5
Energy Kcal/kg	2900	2900	2902	2904	2905
Dry Matter	91.6	91.7	91.8	92.8	92.5
Ash (%)	9.8	9.7	8.7	12.9	8.1
CP (%)	17.8	17.8	17.5	17.3	17.9
EE (%)	6.9	6.1	6.9	8.0	10.6
NDF (%)	32.5	29.5	34.2	34.7	33.8
ADF (%)	11.9	9.5	12.4	12.9	11.7
Calcium (%)	5.4	4.6	4.5	6.6	3.0

Table 4.3 Chemical composition (% DM basis) of layers mash

Layers mash	T1	T2	T3	T4	T5
Energy Kcal/kg	2893	2881	2892	2903	2883
Dry Matter	91.2	91.3	91.3	91.4	91.0
Ash (%)	8.2	7.7	7.2	8.8	7.3
CP (%)	15.4	15.5	15.7	15.9	15.7
EE (%)	4.9	4.7	6.5	6.2	6.8
NDF (%)	18.5	16.7	19.2	19.1	18.9
ADF (%)	4.9	4.6	5.1	4.7	5.1
Calcium (%)	2.8	3.3	3.3	3.2	3.1

The CP level of the diets was formulated to meet the minimum requirement of 20%, 18% and 16% for chicks, grower and layer hens feed respectively. However, differences in CP content among the treatments could be as a result of variability in CP content of raw materials used during feed mixing, most of which were not analysed before use due to machine breakdown. Chick mash had relatively high ether extract (EE) level as seen in T4 (10.3%) and T5 (11.1%) respectively. This could be as a result of high levels of BSFL used during feed mixing as compared to that in other treatments. However, in growers mash, the EE content in feed was quite high for T4 (8.0%) and T5 (10.6%) as compared to the control feed T1 (6.9). Ash content in growers' mash for T4 was highest (12.9%) while T5 had the lowest (8.1%) level. High ash levels in feeds could be as a result of using aldurated FM.

4.2 Amino Acid Profile of Black Soldier Fly larvae and Fishmeal Used in the Experiment

Amino acid composition of BSFLM and FM used in the study is shown in Table 4.4.

Table 4.4 Amino acid composition of BSFLM and FM

Parameter	Content (% DM)	
	BSFLM	FM
CP	43.8	65.7
Methionine	0.6	1.7
Cystine	0.3	0.6
Met. + Cys[a]	0.9	2.2
Lysine	1.9	4.6
Threonine	1.3	2.5
Arginine	1.7	3.6
Isoleucine	1.4	2.5
Luecine	2.4	4.4
Valine	2.0	2.8
Histidine	0.9	1.5
Phenylalanine	1.3	2.4
Glycine	1.8	4.1
Serine	1.4	2.4
Proline	2.1	2.7
Alanine	2.2	3.9
Aspartic acid	3.2	5.6
Glutamic acid	3.1	8.2

[a]Met- Methionine, Cys- Cystine, CP- Crude protein and DM- Dry matter.

Amino acid composition of FM used was superior to that of BSFLM. Also, FM CP content (65.7%) was higher compared to BSFLM CP (43.8%) composition. Crude protein content of BSFLM used in this study was higher than what was reported by De Marco et al. (2015) (36.9% DM) but, slightly higher than Makkar et al. (2014) who reported a CP of (42.1% DM) and similar to what was reported by Onsongo et al. (2018) (43.90% DM). This differences in CP content could be explained by the use of different substrates during larvae growth stage of BSF life cycle. According to Tschirner and Simon (2015) different studies reported that substrate type used during BSF larvae production greatly impact their nutrient composition and total yield. Rearing BSFL on wastes is a self-financing form of reducing pollution, however, the ratios of insects to substrate and favorable conditions for rearing them are not clear (Wang & Shelomi, 2017). The CP content of FM was above the range of 40–50% reported by NRC

(1994), Willis (2003) and Van Eys et al. (2004) and even higher than what was reported by Liebert (2017) and Onsongo et al. (2018) (60.3%). The high CP level obtained means that the FM used was of high quality with minimal human adulteration. High impurity levels in FM is a major problem affecting purchase of this feed ingredient (Kariuki, 2011; Onsongo et al., 2018).

In practical poultry diets, methionine and lysine are the two most limiting amino acids hence the need to add them in their diets (NRC, 1994). Proportions of BSFL amino acids were lower than in FM making it more superior than BSFL. However, the two limiting amino acids; methionine and lysine in BSFL and FM were within the recommended range for exotic chicken (NRC, 1994). The required lysine and methionine for poultry in general is 1.00 and 0.38 g/kg respectively (Boland et al., 2013). However, lysine and methionine levels reported in this study were high meaning used BSFL and FM had good amino acid score. Methionine content of BSFL was lower than that reported by Onsongo et al. (2018) but higher than that reported by Spranghers et al. (2017). However, lysine content of BSFL was similar to that reported by Onsongo et al. (2018) (containing 2.8% lysine and 0.8% methionine). Similar contents for the two limiting amino acids were also reported by Newton et al. (1977) and Onsongo et al. (2018).

4.3 Effect of Substituting Fishmeal with Black Soldier Fly Larvae Meal on Growth Performance of Isa Brown Chicks

Chick final weight, overall weight gain, ADFI, ADG and FCR were significantly different ($P<0.05$) across the treatments. Initial weight was similar for all the five treatments ($P= 0.140$) while final weight was significantly different ($P<0.001$) among the treatments. Chick ADG was significantly different ($P<0.001$) for all the treatments. Overall weight gain was also significant ($P<0.001$) for all the five treatments. Analysis also showed that ADFI was significantly different ($P<0.001$) for all the five treatments, with highest mean observed in T1 of 36.21g while T5 had a lower mean of 24.68g. This implies that increasing levels of insects in layer chick feed leads to a significant decrease in feed intake. Feed conversion ratio was not significantly different ($P<0.054$) for all the treatments (Table 4.5). During the feeding trial period three chicks died from T1 and T3.

Table 4.5 Growth performance of exotic chicks fed five different BSFL-based diets

Parameter	Control	25	50	75	100	P value
Initial weight (g)	96.2±2.6	101.3±1.3	100.9±1.2	102.1±1.3	101.3±1.9	0.140
Final weight (g)	476.2±13.2^{a}	481.5±8.0^{a}	418.0±13.3^{b}	414.7±14.1^{b}	346.7±8.5^{c}	<0.001
Overall weight gain (g)	380.0±14.7^{a}	380.2±8.2^{a}	317.0±14.2^{b}	312.6±14.3^{b}	245.4±8.7^{c}	<0.001
ADG (g)	9.1±0.4^{a}	9.1±0.4^{a}	7.6±0.3^{b}	7.4±0.4^{b}	5.9±0.2^{c}	<0.001
ADFI (g)	36.2±1.7^{a}	33.8±1.5^{a}	29.8±1.3^{b}	27.7±1.3^{bc}	24.7±1.0^{c}	<0.001
FCR	1.8±0.1	1.9±0.1	1.8±0.1	1.9±0.1	1.9±0.1	0.054

$^{a, b, c}$ Means in a row with similar superscript letters are not significantly different at α=0.05. ADG - Average daily gain, ADFI- Average daily feed intake, FCR- Feed conversion ratio, Values = means ± standard error.

These results show that, BSFL meal can substitute FM without any effect on body weight gain, FCR and ADFI, up to 50% in chicks' diets. Growth performance of chicks fed 75% and 100% inclusion level of BSFL meal was poor but, no signs of nutrient deficiencies or higher mortalities were observed. According to Rust (2002) the reason for declining performance of chicks fed on BSFL based diet could be due to the chitin content of BSFL. This component of the invertebrate exoskeleton might have adverse effects on nutrient intake and digestibility in chicks.

Study by Dahiru et al. (2016) established that, for growth performance of spring chicken body weight gain (BWG) in 5%, 7.5% and 10% BSFL inclusion level was highly significant. This means at lower inclusion levels of BSFL chick's growth performance was high compared to higher inclusion levels. However, Awoniyi et al. (2003) and Dahiru et al. (2016) reported negative and insignificant (P>0.05) body weight gain performances at 25% to 100% inclusion levels of insects in poultry diet. Average daily feed intake results differed from Mohammed et al. (2017) findings who stated that feed intake was high for all birds in his treatment groups for the feeding experiment period. Inclusion of fresh BSFL larvae in chicken feed led to improved chicken growth pattern during the rearing period (Moula et al., 2017).

According to Makkar et al. (2014), substituting BSFL for soybean meal in chicks feed resulted to significant weight gain at a rate of 96% compared to the control group containing soybean meal only. However, they only consumed 93% as much feed (Hale, 1973; Makkar et al., 2014; Newton et al., 2005), thus greater FCR of diet containing BSFL meal. Black soldier fly larvae, is a potentially viable alternative to soybean meal and FM in poultry feed and previous research has also identified BSFL as completely viable for other livestock. There is potential to rear insects and raise chicken on BSFL reared on food waste, as this will improve growth performance of such birds (Gaffigan, 2017). According to Nazif and Metekia (2017) and Newton et al. (2005) use of BSFL as component of diet to substitute soybean meal in chicks' diet resulted in good growth and high feed conversion ratio in the birds. This study found that FCR of the chicks was higher for chicks fed BSFLM diets as compared to the control group.

4.4 Effect of Substituting Fishmeal with Black Soldier Fly Larvae on Growth Performance of Exotic Grower Birds

Initial weight of grower (9 weeks old) birds was significantly different ($P<0.001$) across the treatment groups, with birds in T5 having a lower mean live weight than the other four treatments. A significant difference ($P<0.001$) was also observed for final weight where T5 was significantly different from T1, T2 and T3 but not significantly different from T4. Birds in T2 had the highest final mean weight (1264.6g) while T5 had the lightest birds (1049.4g) at the end of the feeding experiment. Overall weight gain was not significantly different ($P=0.260$) for all the five treatments. Average daily weight gain was similar ($P=0.424$) for all the treatments although T2 and T3 had the highest mean values. Birds in T2 had the highest mean overall weight gain of 783.17g although it was not statistically different from other treatments. Average daily feed intake was not significantly different ($P=0.055$) for all the five treatments. Increasing BSFL meal in growers diets led to increased FCR from 0.8 to 0.9. However, no significant difference ($P=0.289$) was observed for all the five treatments. Mortality rate was low at this stage as only 2 birds died from T4. The performance of grower birds is presented in Table 4.6.

Table 4.6: Growth performance of exotic growing hens fed BSFL-based diets at different inclusion levels

Parameter	Control	25	50	75	100	P value
Initial weight (g)	476.2±13.2^{a}	481.5±8.0^{a}	418.0±13.3^{b}	414.7±14.1^{b}	346.7±8.5^{c}	0.001
Final weight (g)	1181.6±29.8^{ab}	1264.6±23.8^{a}	1189.1±30.9^{ab}	1139.6±6.1^{bc}	1049.4±23.5^{c}	0.004
Overall weight gain (g)	705.4±18.9	783.2±19.3	771.0±19.9	724.9±56.6	702.6±21.4	0.260
ADG (g)	9.2±0.5	10.2±0.5	10.0±0.5	9.4±0.5	9.1±0.5	0.424
ADFI (g)	84.6±1.6	84.3±1.5	84.4±1.8	78.3±1.7	75.2±5.7	0.055
FCR	0.8±0.1	0.9±0.1	0.8±0.1	0.8±0.1	0.9±0.1	0.289

$^{a, b, c}$ Means in a row with similar superscript letters are not significantly different at α=0.05. ADG - Average daily gain, ADFI- Average daily feed intake, FCR- Feed conversion ratio, Values = means ± standard error

The main aim of this study was to establish performance of growing birds offered diets with different substitution levels of FM with BSFL. It can be confirmed from the results that, BSFL inclusion in grower diet did not have any significant effect on overall weight gain, ADFI, ADG and FCR. Therefore, BSFL can replace FM up to 100% in grower pullets diet as bird's performance was not significantly different from the birds fed on conventional diet. The findings by Mwaniki et al. (2018) showed an improved feed intake by inclusion of BSFL meal as birds fed 7.5% BSFLM had highest feed intake relative to the control and 5% of defatted BSFL meal. These findings deferred with the present study as ADFI of the grower birds reduced with increased BSFLM in the diets.

According to Widjastuti et al. (2014) quails fed up to 10% BSFL meal in practical diets showed increased feed consumption. Feed intake of 75-84 g/day/bird observed in this study was close to the 63-78 g/day/bird reported by Agbolosu and Teye (2012) but lower than the 130-133 g/day/bird documented by Teye et al. (2012) and Widjastuti et al. (2014) for growing guinea fowls probably due to differences in experimental animals used. An initial mean weight of 346-481 g/bird was recorded in the present study. This could be explained by low feed intake during chick growth phase as birds for the grower phase where sourced from the previous chick feeding experiment. Final mean weight of 1049-1264 g/bird was also recorded during the grower phase. Birds in T5 (100% BSFLM) had low feed intake while the control group had the highest feed intake. Chitin content of pre-pupae exoskeleton might have adverse effect on nutrient digestibility hence contributed to less final weight of the birds in T5. However, Makkar et al. (2014) reported that, substituting soybean meal with BSFL resulted in a significant weight gain but reduced feed intake as compared to the control group thus an improved feed conversion ratio.

Insects' exoskeletons possess chitin which influence feed intake negatively thus interfering with protein utilization in chicken (Józefiak & Engberg, 2015; Longvah et al., 2011). Although chicken are able to produce chitinase in hepatocytes and proventriculus, chitin digestibility is inadequate (Hossain & Blair, 2007; Józefiak & Engberg, 2015), especially in chicks (Ijaiya & Eko, 2009; Józefiak & Engberg, 2015). Black soldier fly larvae meal shows positive effect on birds' weight gain and can be used successfully without fear of any side effects in exotic grower birds. Chitin present in BSF larvae acts like fibre hence used as a prebiotic due to its ability to promote increased intestinal length and weight in birds (Bovera et al., 2015, 2016). These insects contain natural antibiotics thus there is reduced use of antibiotics in birds during rearing and also mortality rate is lower. Black soldier fly larvae are highly palatable to chicken, especially with grower birds as they seek out BSFLM from feeders when feed is introduced rather than continue to feed *ad libitum* feed provided (Ruhnke et al., 2017). As a result, they are a potential partial substitution for FM in poultry feeds, providing added protein with the advantage that they can be reared on the manure from the same birds that consume them thus, recycling waste. Furthermore, BSFL do not necessarily need fortification of some key amino acids (lysine and methionine) for enhanced growth and causes no adverse effects to birds feeding habit as feed intake is not affected (Wang & Shelomi, 2017).

4.5 Growth Performance and Laying Capacity of Layers Fed Different BSFLM-Based Diets

Initial weight, final weight, overall weight gain, ADG, ADFI and FCR were determined and recorded for 25 weeks during which the feeding experiment was conducted. Initial weight of the birds was significantly different (P=0.0065) with birds in T5 being lightest (1504.2g) while those in T2 being heaviest (1701.7g). A significant difference (P=0.0431) was also observed for final weight of the birds with mean weight ranging from 1976.4g to 1722.0g. In terms of overall weight gain, birds in T4 gained more weight (343.9g/bird) as compared to birds in T3 which gained least weight (126.9g/bird). However, ADG was not significantly different (P=0.1348) for all the five treatments during the feeding experiment period. On the other hand, a significant difference (P<0.001) was observed for ADFI for all the treatments with birds in T1 recording the highest ADFI (126.2g/bird) and T5 having the least ADFI (110.9g/bird). Hen day egg production (HDEP) was calculated using the formula; total number of eggs laid on a daily basis divided by number of birds available in the flock on that day multiplied by 100. A statistically significant difference (P<0.001) was observed for HDEP with the highest egg production percentage observed for T2 (75.4) and least egg number observed in T3 (65.5). Birds in T1 (conventional feed) performed similar to T2 and T4 as daily egg production was not significantly different (Table 4.7).

Table 4.7: Growth performance and hen day egg production of exotic layers offered five BSFLM-based diets at different inclusion levels

Parameter	Control	25	50	75	100	P value
Initial weight (g)	1668.3±9.6^{a}	1701.7±28.6^{a}	1680.1±21.7^{a}	1632.5±19.6^{a}	1504.2±52.7^{b}	0.007
Final weight (g)	1863.4±66.5^{ab}	1838.7±37.0^{ab}	1807.0±50.3^{b}	1976.4±46.6^{a}	1722.0±31.0^{b}	0.043
Overall weight gain (g)	195.1±75.8	137.0±67.5	126.9±56.2	343.9±31.3	217.8±48.0	0.135
ADG (g)	4.1±0.4	4.7±0.4	5.1±0.5	5.1±0.4	5.4±0.7	0.352
ADFI (g)	126.2±1.8^{a}	121.4±1.5^{b}	112.5±1.8^{c}	121.1±1.7^{b}	110.9±1.7^{c}	<0.001
FCR	8.6±3.2	6.4±3.2	6.3±2.7	16.2±1.2	11.1±2.1	0.106
HDEP (%)	75.4±1.6^{a}	76.9±1.5^{a}	65.5±1.8^{b}	74.4±1.7^{a}	69.4±1.9^{b}	<0.001

$^{a, b, c}$ Means in a row with similar superscript letters are not significantly different at α=0.05. ADG - Average daily gain, ADFI- Average daily feed intake, FCR- Feed conversion ratio, T- treatment, Values = means ± standard error, HDEP- Hen Day Egg Production

Many published work on exotic laying hens focused on laying performance, weight of egg components and colour, but rarely on egg fatty acid and chemical composition (Agunbiade et al., 2007; Al-Qazzaz et al., 2016; Amao et al., 2010; Makkar et al., 2014; Secci et al., 2018). From the results above, it can be noted that BSFL can substitute FM at different inclusion levels in exotic layer chicken diet without any negative effect on growth performance and laying percentage. Exotic hens can be fed BSFL based feed up to 100% inclusion level with no effect on daily weight gain of the birds. Furthermore, this suggests that BSFL meal can efficiently replace other protein sources, such as soybean meal, groundnut cake and FM up to 75% when formulating layer diet. These findings are in agreement with Barragan-Fonseca et al. (2017) and Maurer et al. (2016) results who reported that half or full substitution of soybean cake by fairly defatted BSFL meal in layers diet had no effect on egg production or feed conversion efficiency, as compared to organic standard diet for layers. However, Dahiru et al. (2016) had different results where a significant difference ($P<0.05$) was reported for final body weight for each treatment. Feed intake appreciably varied ($P<0.05$) among the treatments, which were closely related to their FCR with statistical differences across the entire experimental birds ($P<0.05$) and this matched the findings of the present study. Al-Qazzaz et al. (2016), Decraene (2017) and Jozefiak et al. (2016) reported that FCR was significantly higher with inclusion of BSF larvae. However, Amao et al. (2010) reported that replacing FM with larvae meal by 100g/100g led to a significant increase in the feed conversion ratio of hens.

A significantly ($P<0.001$) highest HDEP was observed in hens diet with 25% BSFL and the least HDEP in hens fed 50% BSFL based diet. Less number of eggs produced by birds in T3 could have been as a result of frequent diarrhoea and lameness shown by the birds during the feeding experiment. The results differ from Al-Qazzaz et al. (2016) who reported that highest HDEP was in the diet of hens fed BSFL at 50%. However, Widjastuti et al. (2014) found out that increasing BSFL levels in quail diets did not have any effect on egg production. Black soldier fly larvae are very palatable to laying hens, as hens are reported to look for BSFL from feeders when feed is introduced rather than continue to eat *ad libitum* feed (Ruhnke, 2017; Wang & Shelomi, 2017). As a result, BSFLM is potential substitution for FM in layer feed, as it has high CP and fat level. These insects have an added advantage that, they can be reared on manure of the same birds that consume them thus, resulting in waste recycling which reduces environmental pollution (Wang & Shelomi, 2017).

4.6 Evaluation of Egg Characteristics of Hens Fed BSFLM-Based Diets

Inclusion of BSFL in layer diet had a significant impact on egg weight, yolk colour, albumen weight and haugh unit parameters (Table 4.8).

Table 4.8: Egg characteristics of exotic layer hens fed five BSFL-based diets at different inclusion levels

Parameter	Control	25	50	75	100	P value
Egg weight (g)	56.6±0.5^{a}	55.2±0.4^{b}	55.1±0.5^{b}	56.9±0.5^{a}	54.4±0.5^{b}	<0.001
Shell thickness (mm)	0.4±0.4	0.4±0.6	0.4±0.6	0.4±0.6	0.4±0.1	0.067
Yolk colour	1.4±0.6^{c}	1.6±0.7^{c}	1.6±0.8^{c}	2.0±0.1^{b}	2.9±0.1^{a}	<0.001
Yolk weight (g)	14.7±0.2	14.6±0.2	14.4±0.2	14.9±0.2	14.6±0.2	0.544
Albumen weight (g)	34.6±0.3^{a}	33.5±0.3^{ab}	33.3±0.3^{ab}	34.3±0.3^{ab}	33.1±0.3^{b}	0.001
Shape index (%)	77.4±0.3	76.7±0.3	77.1±0.4	77.4±0.3	77.9±0.2	0.071
Haugh unit (%)	185.1±0.3^{b}	185.3±0.3^{ab}	185.1±0.4^{b}	186.1±0.4^{a}	183.7±0.3^{c}	<0.001
Yolk index (%)	285.7±3.6	293.0±3.7	296.6±9.2	287.3±3.4	281.1±4.9	0.269

$^{a, b, c}$ Means in a row with similar superscript letters are not significantly different at α=0.05. Values = means ± standard error.

Eggs from T4 were heaviest 56.9g while those from T5 where lightest 54.4g. Egg yolk colour was significantly improved as the level of BSFLM was increased in layer diet and as a result, eggs from T4 and T5 had a much yellow yolk than those from other treatments (25% and 50% BSFLM) and the conventional feed (T1). Shell thickness was not statistically different (P=0.67) for all the treatments as no weak shelled eggs were collected during the entire experimental period. Yolk weight too was not significantly different (P=0.544) for all the five treatments. However, albumen weight was significantly different (P=0.001) with eggs from T1 having the heaviest albumen (34.6g) as compared to those from T3 and T5 which had 33.3g and 33.1g mean albumen weight respectively. Egg shape index and yolk index were not significantly different for all the five treatments. Haugh unit was significantly different (P<0.001) for all the treatments with T4 having the highest percentage (186.1) and the least percentage observed in T5 (183.7).

Chicken eggs are considered human food because they offer a complete balance of essential nutrients such as proteins, vitamins, minerals and fatty acids which are of biological value (Brugalli et al., 1998; Menezes et al., 2012). It also increases consumption of nutritional value foods at a low cost especially for the low-income population (Menezes, 2009; Menezes et al., 2012). Substitution of FM with BSFLM at different inclusion levels did not have any significant difference on shell thickness, egg yolk weight, yolk index and shape index of the egg. However, haugh unit, albumen weight, egg yolk colour and egg weight were influenced by dietary treatments. Productive characteristics such as HDEP, egg mass and egg weight were all influenced by dietary treatments (Table 4.7 and 4.8). Eggs from T4 and T1 were not significantly different in terms of total weight, but eggs from T2, T3 and T5 were lightest. This observation is in agreement with what was observed by Ruhnke et al. (2018) who reported that eggs laid by hens fed on BSFL based feed for 12 weeks were significantly lower in weight than the control group. However, Al-Qazzaz et al. (2016), Decraene (2017) and Ismail et al. (2016) found that egg weight for birds fed 5% BSFL was significantly lower than the egg weight of those fed 1% BSFL meal. These findings differ with the current results as eggs from birds fed 75% BSFL had the highest egg weight as compared to those from the conventional diet although the difference was not significant.

Change in yolk color mostly depends on the concentration, type and ratio of xanthophyll in the feed. Yolk color can also be changed by adaption of the feed ingredients or by adding synthetic and/or natural pigments (Decraene, 2017). However, in this study no pigments or xanthophyll were used but yolk color was changed by more addition of BSFL in layers diets. This means the said insects have natural pigments for yolk color change making it more yellow compared to the conventional feed. A significantly paler egg yolk colour for eggs from birds that fed on BSFL based feed was reported by Ruhnke et al. (2018). Also, Jansen (2018) reported a no significant difference (P>0.05) in yolk colour across all treatments when data for the whole feeding period was analyzed. However, this study findings match with Secci et al. (2018) results who reported that, egg yolk from the BSFL group eggs were significantly redder than the control group. The study also found a clear explanation and confirmation in the total carotenoid content of insect meal and diet containing insect meal (Secci et al., 2018). Therefore, BSFL can replace FM in exotic layer diets without adverse consequences on growth performance, egg production and egg quality parameters.

4.7 Economic Analysis of Using BSFLM-Based Diets in Exotic Layer Hens

Cost benefit ratio (CBR) and return on investment (RoI) were used to calculate economic effect of substituting FM with BSFL in exotic layer diet (Table 4.9).

Table 4.9: Economic analysis of replacing FM with BSFL as alternative protein ingredient in exotic layer diets

	T1	T2	T3	T4	T5	P value
Cost of feed (Kes/kg)						
Layers' mash	46	45	44	43	42	-
Total feed intake (g/hen)						
Entire feeding phase feed intake	1611.3	1520.3	1381.3	1457.2	1307.3	-
Average daily intake	126.2±1.8[a]	121.4±1.5[b]	112.5±1.8[c]	121.1±1.7[b]	110.9±1.7[c]	<0.001
Cost of feed consumed (Kes/hen)						
Layer	1580±0.1[a]	1520±0.1[b]	1410±0.2[c]	1540±0.5[b]	1390±0.2[c]	<0.001
Egg lay (%)	75.4±1.6[a]	76.9±1.5[a]	65.5±1.8[b]	74.4±1.7[a]	69.4±1.9[b]	<0.001
Period-in-lay (days)	175	175	175	175	175	-
Total eggs laid	129	133	113	131	120	-
Sale of eggs (Kes)	1700	1730	1470	1680	1560	-
Gross profit margin	150±0.3[ab]	220±0.2[a]	80±0.3[b]	140±0.4[ab]	190±0.3[a]	0.029
CBR	9±0.3[ab]	14±0.2[a]	6±0.3[b]	9±0.2[ab]	14±0.4[a]	0.041
RoI	920±2.7[ab]	1440±2.3[a]	580±2.7[b]	910±1.9[ab]	1360±3.7[a]	0.041

Cost (Kes/kg) of protein ingredients used in formulating the experimental diets; SBM 90, FM (*omena*) 120, BSFL 85. Means in a row with similar superscript letters are not significantly different at α=0.05, CBR=Cost benefit ratio, RoI=Return on investment.

In this analysis, cost of feed ingredients and sale of eggs at the end of the feeding trial were treated as the only source of costs and profits respectively. Cost of feed, profit margin, CBR and RoI were significantly different ($P<0.001$, $P<0.029$ and $P=0.041$) respectively for the five treatments. The conventional diet (T1) was most expensive costing up to 46 Kes per kg of feed while, T5 was cheapest in terms of total feed cost (41 Kes) per kg of feed and cost of feed consumed (1390 Kes) per bird. Gross profit margin was assumed to be the difference between total cost of feeds and sale of eggs and according to these assumptions birds in T2 and T5 had the highest margins of 220 Kes and 190 Kes respectively. Cost benefit ratio however revealed that birds fed on treatment 2 and 5 had a higher CBR than those from T1 and T3. Calculated ROI on the other hand, showed similar results to CBR in all the five treatments with T2 and T5 recording the highest ROI of 1440 Kes and 1360 Kes respectively.

The price of feed gradually reduced as more BSFL meal was used to substitute FM in layer hens diet resulting in T5 being the cheapest (420 Kes per kg of feed) and T1 most expensive (460 Kes per kg of feed). The results also showed that T3 had the least gross profit margin, CBR and ROI compared to other treatments. However, T2 (220 Kes) and T5 (190 Kes) had the highest gross profit respectively thus BSFL feed was more affordable than the conventional feed. According to Khan et al. (2016) poultry feed costs can be reduced by use of insect meal as a protein source. The results of this study are similar to those reported by Onsongo et al. (2018) with broiler chicken, where partial substitution of soybean and FM with BSFLM resulted in a diet with highest CBR and ROI than the conventional diet. Formulating conventional poultry feed solely depend on FM as the major protein source and this heavily depends on fish importation and overfishing in major lakes in Kenya due to competition with humans for this ingredient (Kolding et al., 2014).

CHAPTER FIVE

CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

This study was conducted to determine the effect of substituting FM with BSFL meal in layer chicks, grower pullets and laying hens' diet on growth performance, laying capacity and egg characteristics. It is therefore concluded that:

(i) Black soldier fly larvae meal inclusion up to 50% in chicks' diet had no adverse effect on ADG, feed intake, body weight gain and FCR.

(ii) Replacement of FM up to 75% in grower and layer diets did not affect ADG, ADFI and hen day egg production.

(iii) Substituting FM with BSFLM in layer diet did not have effect on egg characteristics.

(iv) Use of BSFLM led to a reduction in feed cost of exotic layer chicken to market weight and for maximum egg production under commercial condition.

5.2 Recommendations

Although the results shown are good, the following recommendations are necessary:

(i) Lower levels (up to 50%) of BSFLM can be included in layer chicks' diets for optimum growth.

(ii) In grower and layers' diets, inclusion of BSFLM can be increased up to 75% for optimum performance.

(iii) To achieve maximum laying capacity and good egg characteristics, BSFLM can replace FM up to 75% in exotic layer hens' diet.

(iv) Black soldier fly larvae meal can be used to formulate cheaper and high quality exotic layer diets.

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APPENDICES

Appendix 1: ANOVA Tables Showing Growth Performance of Chicks

Feed intake

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	4746	1186.4	10.91	3.05e-08 ***
Residuals	283	30765	108.7		

Average daily weight gain

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	392	98.01	13.32	5.89e-10 ***
Residuals	283	2083	7.36		

Initial weight

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	201	50.26	1.835	0.14
Residuals	43	1178	27.40		

R-Square	Coeff Var	Root MSE	Initial weight Mean
0.145781	5.221004	5.234190	100.2526

Final weight

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	111668	27917	20.91	<0.001
Residuals	43	57411	1335		

R-Square	Coeff Var	Root MSE	final weight Mean
0.660448	8.535958	36.53966	428.0675

Overall weight gain

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	115255	28814	19.4	<0.001
Residuals	43	63880	1486		

Feed conversion efficiency (FCE)

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	0.3001	0.07502	2.686	0.0438
Residuals	43	1.2009	0.02793		

Appendix 2: ANOVA Output for Economic Analysis

Feed cost mean

Source	DF	Squares	Mean Square	F Value	Pr > F
Model	4	8.50403238	2.12600809	40.05	<.0001
Error	10	0.53089435	0.05308944		
Corrected Total	14	9.03492673			

R-Square	Coeff Var	Root MSE	feed cost Mean
0.941240	1.551200	0.230411	14.85375

Gross profit margin

Source	DF	mean sum of Squares	Mean Square	F Value	Pr > F
Model	4	3.22118542	0.80529636	4.26	0.0288
Error	10	1.89130847	0.18913085		
Corrected Total	14	5.11249390			

R-Square	Coeff Var	Root MSE	profit Mean
0.630061	28.17423	0.434892	1.543580

Cost benefit Ratio mean

Source	DF	mean sum of Squares	Mean Square	F Value	Pr > F
Model	4	0.01502429	0.00375607	3.73	0.0414
Error	10	0.01005719	0.00100572		
Corrected Total	14	0.02508149			

R-Square	Coeff Var	Root MSE	CBR Mean
0.599019	30.43084	0.031713	0.104214

Return on investment mean

Source	DF	Squares	Mean Square	F Value	Pr > F
Model	4	150.2429348	37.5607337	3.73	0.0414
Error	10	100.5719477	10.0571948		
Corrected Total	14	250.8148825			

R-Square	Coeff Var	Root MSE	ROI Mean
0.599019	30.43084	3.171308	10.42136

Appendix 3: ANOVA Outputs for Grower Birds

Average daily weight gain

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	101	25.37	0.97	0.424
Residuals	523	13685	26.17		

R-Square	Coeff Var	Root MSE	ADG Mean
0.005539	47.81067	4.690353	9.810263

Daily feed intake

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	7769	1942.2	2.335	0.0546
Residuals	523	435004	831.7		

R-Square	Coeff Var	Root MSE	Feed Intake Mean
0.017545	35.42960	28.84003	81.40097

Initial weight

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	111668	27917	20.91	1.25e-09 ***
Residuals	43	7411	1335		

Final weight

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	234152	58538	4.543	0.00378
Residuals	43	554101	12886		

Overall weight gain

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	53793	13448	1.369	0.26
Residuals	43	422337	9822		

R-Square	Coeff Var	Root MSE	overall weight gain Mean
0.112980	13.41375	99.10492	738.8308

FCE

Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)
Treatment	4	0.0680	0.01700	1.289	0.289
Residuals	43	0.5671	0.01319		

R-Square	Coeff Var	Root MSE	FCE Mean
0.107067	13.83847	0.114836	0.829829

Appendix 4: ANOVA Outputs for Layer Birds

ADG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	81.724663	20.431166	1.11	0.3522
Error	370	6819.246099	18.430395		
Corrected Total	374	6900.970762			

R-Square	Coeff Var	Root MSE	ADG Mean
0.011842	88.15002	4.293064	4.870179

Daily feed intake

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	12580.37053	3145.09263	14.41	<.0001
Error	370	80748.35143	218.23879		
Corrected Total	374	93328.72196			

R-Square	Coeff Var	Root MSE	ADFI Mean
0.134796	12.47547	14.77291	118.4156

Initial weight

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	74031.6755	18507.9189	6.80	0.0065
Error	10	27220.8005	2722.0801		
Corrected Total	14	101252.4760			

R-Square	Coeff Var	Root MSE	Initial weight mean
0.731159	3.186405	52.17356	1637.380

Final weight

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	102452.8961	25613.2240	3.68	0.0431
Error	10	69626.8045	6962.6805		
Corrected Total	14	172079.7006			

R-Square	Coeff Var	Root MSE	Final weight Mean
0.595380	4.531205	83.44268	1841.512

Overall weight gain

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	102452.8961	25613.2240	3.68	0.0431
Error	10	69626.8045	6962.6805		
Corrected Total	14	172079.7006			

R-Square	Coeff Var	Root MSE	Final Mean
0.595380	4.531205	83.44268	1841.512

FCE

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	204.3544820	51.0886205	2.53	0.1063
Error	10	201.7017663	20.1701766		
Corrected Total	14	406.0562482			

R-Square	Coeff Var	Root MSE	FCE Mean
0.503266	46.26279	4.491122	9.707849

Number of eggs

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	16497.9731	4124.4933	7.80	<.0001
Error	905	478660.8059	528.9070		
Corrected Total	909	495158.7790			

R-Square	Coeff Var	Root MSE	number of eggs mean
0.033319	31.79927	22.99798	72.32234

Appendix 5: ANOVA Outputs for Egg Characteristics

Egg weight

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	594.21338	148.55335	4.83	0.0008
Error	645	19834.49777	30.75116		
Corrected Total	649	20428.71115			

R-Square	Coeff Var	Root MSE	egg weight Mean
0.029087	9.967487	5.545373	55.63462

Egg shape index

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	103.691052	25.922763	2.17	0.0708
Error	645	7701.469729	11.940263		
Corrected Total	649	7805.160780			

R-Square	Coeff Var	Root MSE	shape index Mean
0.013285	4.470192	3.455469	77.30022

Egg yolk colour

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	177.301538	44.325385	27.61	<.0001
Error	645	1035.500000	1.605426		
Corrected Total	649	1212.801538			

R-Square	Coeff Var	Root MSE	yolk colour Mean
0.146192	67.12186	1.267054	1.887692

Egg yolk index

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	19824.009	4956.002	1.30	0.2689
Error	645	2460204.084	3814.270		
Corrected Total	649	2480028.093			

R-Square	Coeff Var	Root MSE	yolk index Mean
0.007993	21.39013	61.75978	288.7303

Egg yolk weight

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	16.789323	4.197331	0.77	0.5438
Error	645	3508.182077	5.439042		
Corrected Total	649	3524.971400			

R-Square	Coeff Var	Root MSE	yolk weight Mean
0.004763	15.93234	2.332175	14.63800

Shell thickness

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	0.03889169	0.00972292	2.21	0.0666
Error	645	2.83938231	0.00440214		
Corrected Total	649	2.87827400			

R-Square	Coeff Var	Root MSE	shell thickness Mean
0.013512	16.57887	0.066349	0.400200

Albumin weight

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	230.160644	57.540161	4.58	0.0012
Error	645	8103.902222	12.564189		
Corrected Total	649	8334.062866			

R-Square	Coeff Var	Root MSE	albumin weight Mean
0.027617	10.49649	3.544600	33.76940

Haugh unit

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	4	399.68396	99.92099	6.66	<.0001
Error	645	9680.78759	15.00897		
Corrected Total	649	10080.47155			

R-Square	Coeff Var	Root MSE	haugh unit Mean
0.039649	2.093550	3.874142	185.0513

Appendix 6: List of Plates

Plate 1. Adult black soldier flies

Appendix 7: Researh Pictorial

Plate 2. Black Soldier Fly Larvae used to Formulate Experimental Feeds

Experimental chicks in a brooder

Egg quality determination

Experimental grower pullets at 18 weeks

Experimental layer hens at 30 weeks of age

Taking weights of experimental birds

Vaccination schedule used during feeding trial.   Yolk colour chart used during egg analysis

Appendix 8: Publications and Conference Presentation

i. Substituting fishmeal with black soldier fly larvae on performance and carcass characteristics of exotic layer chicks and grower pullets. Animal Production Society of Kenya, April 2019 Annual Scientific Symposium *(Presented).*

ii. Substitution of Fishmeal by Black soldier fly larvae on performance, laying percentage and egg quality of Isa Brown layers (*Under review*).

Appendix 9: Published Manuscript Abstract

sustainability MDPI

Article

Cost-Effectiveness of Black Soldier Fly Larvae Meal as Substitute of Fishmeal in Diets for Layer Chicks and Growers

Esther Khayanga Sumbule [1,2], Mary Kivali Ambula [2], Isaac Maina Osuga [3], Janice Ghemoh Changeh [4], David Miano Mwangi [5], Sevgan Subramanian [1], Daisy Salifu [1], Peter A. O. Alaru [5], Macdonald Githinji [5], Joop J. A. van Loon [6], Marcel Dicke [6] and Chrysantus M. Tanga [1,*]

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Abstract: The acceptance of eco-friendly black soldier fly larvae meal (BSFLM) as sustainable alternative protein ingredient in poultry feeds continues to gain momentum worldwide. This study evaluates the impact of BSFLM in layer chick and grower diets on the growth, carcass quality and economic returns. Mean weekly weight gain and total live weight per chick and grower varied significantly. The highest final weight gain was achieved when birds were provided diet with 25.6% BSFLM. Average daily feed intake (ADFI), average daily weight gain (ADG) and overall weight gain of the chick varied significantly, except for the feed conversion ratio (FCR). For grower birds, ADFI, ADG, FCR and overall weight gain did not vary significantly across the various feeding regimes. The weight of the wings and drumsticks had a quadratic response with a maximum weight obtained at 33% inclusion of BSFLM. The weight of the internal organs were not significantly affected by dietary types. Positive cost–benefit ratio and return on investment was recorded for diet types with higher BSFLM inclusion levels (>75%). Diets with 25% and 100% BSFLM inclusion were the most suitable and cost-effective, respectively. Thus, BSFLM represents a promising alternative source of protein that could be sustainably used in the poultry industries.

Keywords: black soldier fly; insect-based feed formulation; chicken layers; carcass and organs yield; profitability; sustainable intensification

Appendix 10: NACOSTI Research Permit

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