EFFECT OF DRIED BLACK SOLDIER FLY (*HERMETIA ILLUCENS*) LARVAE FEED ON THE BLOOD PARAMETER, CARCASS QUALITY AND COST ANALYSIS OF CROSSBREED VILLAGE CHICKEN

By

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A RESEARCH PAPER SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE DEGREE OF DOCTOR OF VETERINARY MEDICINE

FACULTY OF VETERINARY MEDICINE

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2025

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ABSTRACT

An abstract of the research paper presented to the Faculty of Veterinary Medicine, Universiti Malaysia Kelantan, in partial requirement of the course DVT 55204 – Research Project.

The poultry industry is an important component of Malaysia's livestock sector yet rising feed costs have increased interest in sustainable alternative protein sources. Black Soldier Fly Larvae (BSFL) offers a promising option due to their high nutritional value and potential cost efficiency. This study evaluated the effects of dried BSFL supplementation on blood parameters, carcass characteristics, and feed cost in crossbred village chickens (Ayam Kampung Kacuk) over a 9-week (63-day) period. Thirty chickens were assigned to three dietary treatments: T1 (control commercial diet), T2 (commercial diet + 5% dried BSFL), and T3 (commercial diet + 10% dried BSFL). Growth performance, feed intake, blood parameters, and carcass traits were assessed. Overall, BSFL supplementation did not affect selected blood parameters, with all values remaining within normal physiological ranges. While final body weight and major carcass components showed no significant differences among treatments, certain parts including head, thigh, and heart weights displayed significant variation. Feed cost analysis suggested that BSFL inclusion may influence cost efficiency without negatively impacting growth or carcass yield. These findings indicate that BSFL can be used as a partial feed ingredient in village chicken diets without adverse effects on performance or health.

Keywords: Commercial concentrate diet, Cost analysis, Dried black soldier fly larvae, Growth performance, Carcass quality, Blood parameter and Feed cost analysis.

ABSTRAK

Abstrak daripada kertas penyelidikan yang dibentangkan kepada Fakulti Perubatan Veterinar, Universiti Malaysia Kelantan, sebagai keperluan sebahagian daripada kursus DVT 55204 – Projek Penyelidikan.

Industri ternakan ayam merupakan komponen penting dalam sektor ternakan Malaysia, namun kenaikan kos makanan telah meningkatkan minat terhadap sumber protein alternatif yang stabil. Larva Lalat Tentera Hitam (Black Soldier Fly Larvae, BSFL) menawarkan pilihan yang menjanjikan kerana nilai pemakanan yang tinggi dan potensi penjimatan kos. Kajian ini menilai kesan penambahan BSFL kering terhadap parameter darah, ciri-ciri karkas, dan kos makanan dalam ayam kampung kacuk sepanjang tempoh 9 minggu (63 hari). 30 ekor ayam dibahagikan kepada tiga rawatan diet: T1 (diet komersial sebagai kawalan), T2 (diet komersial + 5% BSFL kering), dan T3 (diet komersial + 10% BSFL kering). Prestasi pertumbuhan, pengambilan makanan, profil darah, dan ciri-ciri karkas dinilai. Secara keseluruhan, penambahan BSFL tidak menjejaskan parameter darah terpilih, dengan semua nilai berada dalam julat fisiologi normal. Walaupun berat badan akhir dan komponen utama karkas tidak menunjukkan perbezaan yang signifikan antara rawatan, beberapa bahagian seperti berat kepala, peha, dan jantung menunjukkan perbezaan yang signifikan. Analisis kos makanan mencadangkan bahawa penambahan BSFL mungkin mempengaruhi kecekapan kos tanpa menjejaskan pertumbuhan atau hasil karkas. Penemuan ini menunjukkan bahawa BSFL boleh digunakan sebagai sebahagian daripada diet ayam kampung tanpa kesan negatif terhadap prestasi atau Kesihatan.

Kunci: Diet konsentrat komersial, Analisis kos, Larva Lalat Tentera Hitam kering, Prestasi pertumbuhan, Kualiti karkas, Parameter darah, Analisis kos makanan.

CERTIFICATION

This is to certify that we have read this research paper entitled **'Effect of Dried Black Soldier Fly (*Hermetia Illucens*) Larvae Feed on the Blood Parameter, Carcass Quality and Cost Analysis of Crossbred Village Chicken'** by Lokman Bin Jasni, and in our opinion it is satisfactory in terms of scope, quality and presentation as partial fulfillment of the requirement for the course DVT 55204 Research Project.

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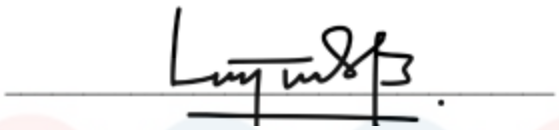

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Family

Thank You

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ABBREVIATION

ALP	-	Alkaline Phosphatase
ALT	-	Alkaline Transferase
ANOVA	-	Analysis Varian
AWG	-	Average Weekly Gain
BSF	-	Black Soldier Fly
BSFL	-	Black Soldier Fly Larvae
BUN	-	Blood Urea Nitrogen
BW	-	Body Weight
BWG	-	Body Weight Gain
CF	-	Crude Fat
CFr	-	Crude Fibre
CK	-	Creatine Kinase
CP	-	Crude Protein
DBSFL	-	Dried Black Soldier Fly Larvae
DM	-	Dry Matter
DOC	-	Day-old chick
FCR	-	Feed conversion ratio
FI	-	Feed intake
g	-	gram

kg	-	kilogram
LDH	-	Lactate Dehydrogenase
PCV	-	Packed Cell Volume
RM	-	Ringgit Malaysia
SEM	-	Standard Error Mean
T1	-	100% commercial concentrate diet
T2	-	5% DBSL, 95% commercial concentrate diet
T3	-	10 DBSFL, 90% commercial concentrate diet
UMK	-	Universiti Malaysia Kelantan

CHAPTER 1

1.0 INTRODUCTION

In Malaysia, the poultry industry is regarded as one of the most important livestock industries that contributed to the largest share of gross output value in the livestock sub-sector at 89.9% valued at RM16.8 billion in 2021 (Department of Statistics Malaysia, 2023). According to the Department of Veterinary Services (2012), Malaysia has fully achieved self-sufficiency level in the poultry industry since 1984.

Poultry meat is one of the crucial sources of high-quality proteins, minerals and vitamins to balance the human diet (Ravindran, 2009). The rising demand for poultry meat, driven by population growth and nutritional needs, exerts significant pressure on the efficiency of the poultry sector. In fact, Malaysia supplies 81% of protein source for the domestic market. According to the Department of Veterinary Services (2014), 1.8 million broiler chickens are produced per day in Malaysia.

However, due to many factors such as COVID 19, conflict between Russia and Ukraine, and internal country political issues. Our country faces a chicken shortage problem. It is necessary to develop a long-term strategy to improve food security and ensure the cost of living is reasonable (BERNAMA, 2022). Conventional feed ingredient diets such as maize, fish meal, meat and bone meal, soybean meal, groundnut cake and others are becoming competitive and expensive in developing countries. Hence, the search for alternative feed sources has become inevitable to reduce the feed cost (Swain et al., 2014).

According to Ongsogo et al. (2018) Black Soldier Fly Larvae (BSFL) can replace up to 15% of the conventional feed ingredients in broiler diets reducing the feed cost by 19% when being compared to Kenyan soybean and fish meal diets by indicating that BSFL may be already a cheaper ingredient option with no adverse effects in broiler production parameters in some countries. Black Soldier Fly Larvae (BSFL) known as *Hermetia illucens* larvae. It is a saprophytic insect that were fed on organic waste such as plant residues, animal manure, and waste, food waste, agricultural byproducts, or straw (Nguyen, 2015). Its habitat can be found widely in warm temperatures at the tropical and subtropical areas (Guo et al., 2021).

1.1 RESEARCH PROBLEM STATEMENT

The increasing demand for animal proteins, driven by a growing worldwide population and enhanced living standards in developing nations, has escalated the costs of material used in animal feed, such as fishmeal, fish oil, soybean meal, and cereals. The global economic downturn has made this problem become more challenging, so the poultry industry needs to find new ways to meet social, environmental, and economic demands. Therefore, growth promoters are usually incorporated into the broiler diets to boost farm performance and profits. Black soldier fly larvae (BSFL) have become a promising nutrient substitute as they contain high protein, lipid levels, and abundant vitamins and minerals content similar to soybean or fishmeal. By partially substituting other protein sources, BSFL has the potential to reduce the cost of chicken feed compositions and improve carcass characteristics as good carcass quality is the aim of the farmers to market their product. Additionally, the meal should not have any negative impacts on the chicken's blood parameters, which could degrade the quality of their meat. This is one of the main arguments in favor of using insects like BSFL as a substitute source

of protein in animal feed. Therefore, the goal of this project is to evaluate the blood parameter, carcass quality and feed costs of crossbred village chickens fed dried BSFL.

1.2 RESEARCH QUESTION

1. What is the effect of Dried Black Soldier fly larvae (DBSFL) on blood parameters of crossbred village chicken?

2. How does the Dried Black Soldier Fly Larvae (DBSFL) improve the carcass quality towards the crossbred village chickens?

3. What is the cost analysis of feeding crossbred village chicken with a new combination fed with dried Black Soldier Fly Larvae (BSFL) for 9 weeks compared to the commercial feeding?

1.3 RESEARCH HYPOTHESIS

H0: Dried Black Soldier Fly larvae do not have any influence on blood parameters of crossbred village chicken.

H1: Dried Black Soldier Fly larvae will have an influence on the blood parameters of crossbred village chicken.

H0: Dried Black Soldier Fly larvae do not have any influence on the carcass quality of crossbred village chicken.

H1: Dried Black Soldier Fly larvae will have an influence on the carcass quality of crossbred village chicken.

H0: There is no cost analysis difference when feeding chicken with Dried Black Soldier Fly larvae compared to the commercial feed diet.

H1: There is a cost analysis difference when feeding chicken with Dried Black Soldier Fly larvae compared to the commercial feed diet.

1.4 RESEARCH OBJECTIVE

1. To compare the blood parameters between crossbred village chickens that had been fed with Dried Black Soldier Fly Larvae and the control group.

2. To determine the effect of Dried Black Soldier Fly Larvae (BSFL) on the carcass quality in crossbred village chickens.

3. To determine the feed cost analysis of crossbred village chicken after introducing a new combination of dried Black Soldier Fly Larvae (BSFL).

CHAPTER 2

2.0 LITERATURE REVIEW

Livestock products were sold to Malaysian consumers as a protein source such as cattle, chicken, pig and others. In 2021, the poultry industry contributed the largest share of gross output value in the livestock sub-sector at 89.9% valued at RM 16.8 billion, surpassing other livestock, which were swine, cattle and buffaloes (DOSM, 2023). Department of Veterinary Services (2012) reported that the Malaysian broiler industry has been self-sufficient in broiler production since 1984. However, the popularity of chicken as a cheaper source of protein among Malaysians may be affected by the increasing production cost of chicken, especially feed costs (Bahri et al., 2019; Shaban & Alabboodi, 2019).

In addition, black soldier fly larvae (BSFL) have a high protein content and are utilized as a feed supplement for livestock, primarily pigs and poultry. This formulation was tested in the first poultry species which is quail. As a result, BSFL showed a high feed intake and enhanced feed conversion ratio (FCR) (Widjastuti et al., 2014). In some studies, adding 5% to 15% BSFL to feed can save feed costs while simultaneously enhancing health and growth performance. However, there is a significant lack of information in the literature about how BSFL affects the quality of the carcass, the assessment of blood parameters, and feeding cost analysis in rearing crossbred village chickens in Malaysia.

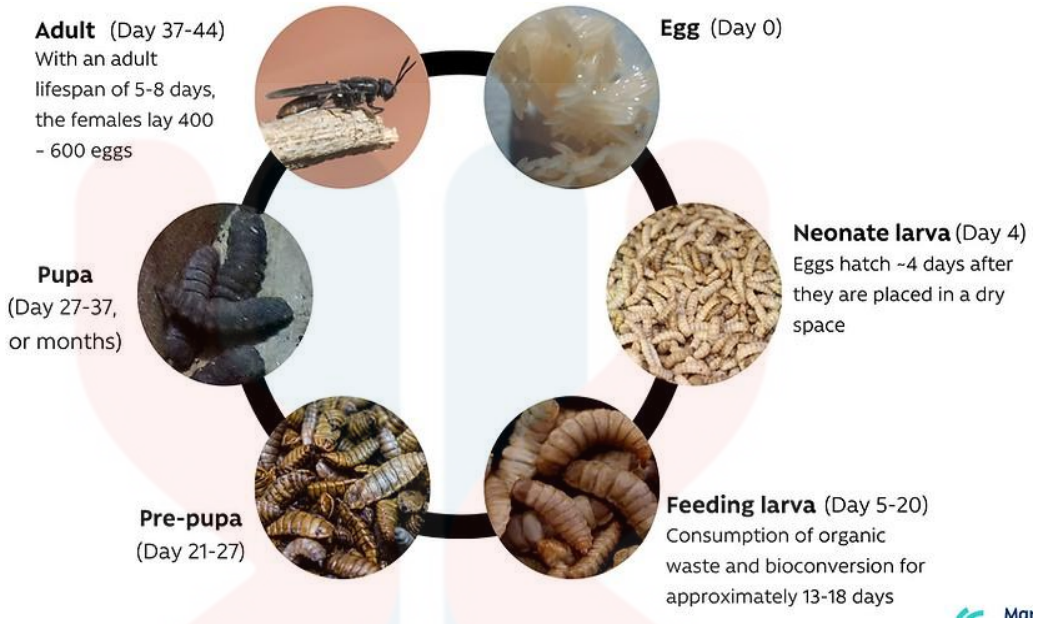

Figure 1: Life cycle of Black Soldier Fly (BSF) *Hermetia illucens.*

Figure 1 shows the Black Soldier Fly (BSF) life cycle which has five main stages including eggs, larvae, prepupae, pupae, and adult flies (Smets et al., 2020) (Figure 1). However, larva and pupal stage is the highest nutrition and large, but it depends on the quality of food with 18% to 33% fat and 32% to 53% protein (Chippindale et al., 2004). The life span of BSFL approximately within 20 to 22 days with the pupa on 6 to 8 days while the adult metamorphosis for the last 14 days (Singh A et al., 2019). In fact, the BSFL does not serve as vectors for disease transmission (Sheppard et al., 2002).

The BSFL is characterized by a high food conversion rate (FCR), short reproductive cycle, and high content of fat, protein, minerals, and vitamins (El-Hack et al., 2020). The feed conversion rate (FCR) of BSFL previously reported has been better than mealworms and crickets, and its survival rate, the composition of nitrogen and phosphorus do not change greatly with a change in diet (Oonincx et al., 2015). Previous study also reported that diets supplemented

with compared to other protein feeds including fishmeal, fish oil, soybean meal, and cereals (Veldkamp, 2015).

The birds' health status is revealed by the serum biochemical markers. The study's findings demonstrated that the broiler chickens were within physiological parameters and that the fats from BSF and MBSF larvae had no effect on the animals' overall health. The findings concur with those of Schiavone et al. (2019) and Sypniewski et al. (2020) who found no discernible differences in the blood parameters of broiler chickens and turkey poults fed fat from BSF larvae. In contrast to coconut oil, Kim et al. (2020) demonstrated that BSF larvae fat reduced total cholesterol and HDL cholesterol in serum samples. However, serum levels of triglycerides, uric acid, ALT, and AST were unaffected by BSF larvae fat.

Black soldier fly larvae (BSFL) are rich in essential amino acids, such as lysine and methionine, which are important for poultry production (Spranghers et al., 2017). The meat quality of chickens fed BSF has been similar to that of the chicken fed commercial diet, therefore, they can be used as a partial substitution for soybean meal (Pieterse et al., 2019). Thus, BSFL could be used as a partial replacement for chicken feed by providing additional protein in addition to being raised on chicken excrement.

The use of Black Soldier Fly Larvae (BSFL) as an alternative protein source in broiler chicken diets has shown significant potential in reducing feed costs, which constitute the largest portion of poultry production expenses. BSFL are rich in essential amino acids and fats, and can be produced sustainably using organic waste, lowering dependency on more expensive

conventional feed ingredients like soybean meal and fishmeal. Studies have demonstrated that partially replacing traditional protein sources with BSFL meals can maintain or even improve broiler growth performance while significantly reducing overall feed costs (Mwaniki et al.,2020). This makes BSFL a promising solution for enhancing the economic viability of poultry farming.

CHAPTER 3

3.0 METHODOLOGY

3.1 Location

The experiment was conducted at the Ladang Pengajar dan Keusahawanan Veterinar, Fakulti Perubatan Veterinar, Universiti Malaysia Kelantan, Bachok, Malaysia (5°59'35.8"N102°24'10.1E") (Figure 2).

Figure 2: Ladang Pengajar dan Keusahawanan, FPV, UMK

3.2 Experimental design

Thirty crossbred village chickens were used for the experiment where the animals were divided into three groups (n=10) of crossbred village chickens. The vaccinated day-old chicks were fed a starter diet from day 1 to day 21 and a grower diet from day 22 to day 63 (Figure 3). The starter and grower feeds for three groups were added with dried BSFL.

Figure 3: Preparation cage for A: day 1 to day 21 and B: day 22 to day 63.

The animals were randomly distributed among three treatments (Table 1) which are treatment 1 (T1) for the control 100% commercial concentrate diet, treatment 2 (T2) for 5% BSFL and 95% commercial concentrate diet followed by treatment 3 (T3) for 10% BSFL and 90% commercial concentrate diet.

Table 1: The ratio of dried BSFL used in different treatments.

Treatment (T)	**Black Soldier Fly Larvae (BSFL)**	**Commercial Concentrate Diet**
T1	-	100%
T2	5%	95%
T3	10%	90%

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet)

3.3 Sampling Method and Procedure

3.3.1 Sample Preparation

The commercialized diet and dried Black Soldier Fly Larvae (BSFL) will be purchased from HK Leong Sdn Bhd and Bioloop Sdn Bhd, respectively. The dried BSFL will be air-dried and sun-dried to remove excess water residue from the surface. The dried BSFL was ground into smaller pieces by using a grinder. Moderate coarse texture powder is made and stored in the container. However, it must be stored in a dry area at room temperature and kept away from direct sunlight. The basal diet used in this experiment is from the commercial concentrate diet broiler feed from HK leong Sdn Bhd and Bioloop Sdn Bhd is 201C starter feed from day 1 to day 21 and 202P grower feed from day 22 to day 63. The commercial concentrate diet starter and grower feed were added with dried BSFL powder into two rations, as shown in Table 1. All preparations were done in the Animal Production Lab, FPV, UMK.

Figure 4: 201 Starter (Left) and 202P Grower (Right)

Source: Gold Coin Feedmills (M) Sdn. Bhd.

Table 2: Nutrition composition of the commercial concentrate diet and dried BSFL.

Parameter	201C Starter	202P Grower	DBSFL
Moisture (%)	13.0	13.0	1.16
Ash (% DM)	8.0	8.0	13.9
Crude Protein (% DM)	21.0	19.0	44.1
Crude Fiber (% DM)	5.0	5.0	6.69
Crude Fat (% DM)	4.5	5.0	24.3

Source: Gold Coin Feedmills (M) Sdn. Bhd, HK leong Sdn Bhd and Bioloop Sdn Bhd.

Figure 5: The commercial concentrate diet, 201c starter feed

Figure 6: The commercial concentrate diet, 202p grower feed

Figure 7: The dried BSFL before grinding into moderate coarse texture powder.

3.3.2 Proximate Analysis

The material and ingredients of commercialized starter, grower and dried BSFL was bought from local suppliers (HK leong Sdn Bhd and Bioloop Sdn Bhd). Each sample (150g) was used to determine moisture, dry matter, ash, crude protein, crude fibre, and crude fat for the broiler feeds that were performed by using the methods and formulas according to the Association of Official Analytical Chemists (AOAC) (2007). The sample are:

a. Commercial Concentrate Diet (Starter)
b. Commercial Concentrate Diet (Grower)
c. Dried Black Soldier Fly Larvae (BSFL)
d. Commercial Concentrate Diet (Starter) mix with 5% dried BSFL
e. Commercial Concentrate Diet (Grower) mix with 5% dried BSFL
f. Commercial Concentrate Diet (Starter) mix with 10% dried BSFL
g. Commercial Concentrate Diet (Grower) mix with 10% dried BSFL

3.3.2.1 Determination of Moisture and Dry Matter

Use an oven to dry the porcelain crucible at 105 °C for 30 minutes and cold with desiccators for 30 minutes. Then, weigh the dried empty porcelain crucible. Next, weigh the 3g of feed samples that are put inside the crucible (W1) and by using an oven, dried at 105 °C for 12 to 24 hours. This crucible will be cold inside the desiccator and reweighed (W2). The dry matter and moisture of sample will be calculated according to the formula, Moisture (%) =

$$[(W1-W2) / W2] \times 100$$

Information:

W1 = Weight of crucible and sample before drying

W2 = Weight of crucible and sample after drying

Dry Matter (%) = 100 – Moisture (%)

3.3.2.2 Determination of Ash

The dried sample from the dry matter analysis was used for the determination of ash. Use an oven to dry the porcelain crucible at 105 °C for 30 minutes and cold with desiccators for 30 minutes. Then, weigh the dried empty porcelain crucible (W1). Before ash was at 550°C for 4 hours, dried samples inside the crucible were weighed (W2). Next, it will be cold in the desiccator before weighing (W3). The ash (%) will be calculated using the formula as follows, Ash (%) =

$$[(W2-W1) / W3 \times 100]$$

Information:

W1= Weight of empty crucible

W2= Weight of sample

W3= Weight of crucible and sample after ashing

3.3.2.3 Determination of Crude Protein

The Kjeldahl method will be used to determine the crude protein. It consists of 3 main stages which are digestion with sulphuric acid, distillation and titration. Digestion with sulphuric acid as the first stage used 0.5g sample together with 12mL of 98% sulphuric acid (H_2SO_4) and added a Kjeldahl catalyst tablet into a Kjeldahl tube with slow heat for 4 hours. Then, 3 mL hydrogen peroxide (H_2O_2) will be added to the mixture and boiled at 270°C for an hour, then increased to 400 °C for another hour. After the digestion of samples is complete, the flasks will be left to be cold for 40 minutes.

By adding 32% of sodium hydroxide (NaOH) inside the digestion flask that connected to the Erlenmeyer receiving flask, consisting of 75 mL of 2% boric acid and eight drops of methyl red and bromocresol green indicators for the distillation process.

Ammonium borate was formed from the end product of distillation. Then, titrate with 0.1 M hydrochloric acid (HCl) until a pink solution is observed. The volume and concentration of HCl used in the titration stage will be calculate to determine the number of mol of nitrogen in the sample. Crude protein content (%) will be expressed as nitrogen content multiplied by 6.25. Percentages of nitrogen and crude protein will be calculate using the following formula, Nitrogen (%) =

[(Volume of titrant-Blank value) × acid normality × 1.4007 / Weight of sample (g)]

Crude protein (%) = Nitrogen (%) × 6.25

Information:

Blank value= 0.2 mL

Acid normality= 0.1 mL

Protein factor= 6.25

3.3.2.4 Determination of Crude Fibre

To determine the crude fibre, Gerhardt Fibretherm was used by using H_2SO_4 and NaOH to digest the feed sample. The oven was used for 1 hour to dry the empty fibre bags and weight (W1). This was followed by, 30 minutes for the crucible in the furnace, then cold using a desiccator by transferring in it and weight (W6). 1g of sample feed was weighed and put into the fibre bags that contained glass spacer (W2) to be put into the carousel. Later, the carousel with fibre bags was placed in a beaker containing 360 mL of 0.13 mol/L H2SO4. The heating plate of the Gerhardt Fiber Bag System apparatus will be used to place the beaker. At the highest temperature of boiling, slowly and gently reduce within 30 minutes. Remove the beaker from the heating plate and rinse with hot water on the carousel and fibre bag.

Use 360 mL of 0.313 mol/L NaOH to pour into the beaker and carousel with the fibre. Tap water was used to rinse the fibre bags and dried using fibre-free tissue.

The fibre bags will dry by leaving overnight in the oven at 105°C. Then, weigh (W3) the crucible by placing the dried fibre bags and placing them in the furnace at 550 °C for 4 hours. Next, place in the desiccator by removing the crucible and fibre bags from the furnace and weigh (W4). Blank value (W5), the crucible (W6) as mentioned before and the crucibles and ash of the empty fibre bags (W7). The formula is used to determine crude fiber percentage. Crude fibre (%) =

$$[(W3-W1)-(W4-W5) / W2] \times 100$$

Information:

W1 = Weight of fibre bag (g)

W2 = Weight of sample (g)

W3 = Weight of crucible with fibre bag after digestion (g)

W4 = Weight of crucible and ash (g)

W5 = Weight of blank value of the empty fibre bag (g), (W7 – W6)

3.3.2.5 Determination of Crude Fat

By using an oven, dried the round-bottomed flask to remove the moisture and cold with a desiccator before weighing (W1). The samples of 3g feed were added into the extraction thimble, covered with cotton wool and placed inside the Soxhlet apparatus. The round-bottomed flask was added with 250mL of petroleum benzene and fixed with Soxhlet heating plate. Then, the condenser was attached to the Soxhlet apparatus, and the tap water was opened. The oven will open overnight at 80°C for the round-bottomed flask to be dry. The flasks were taken out of the oven and let cooled in the desiccators to be weighed (W3). The crude fat (%) for all the feed samples was calculated based on the following formula, Crude fat (%) =

$$[(W3-W2) / W1 \times 100]$$

Information:

W1= Weight of empty round-bottomed flasks

W2= Weight of sample

W3= Weight of dried round-bottomed flasks after evaporation

3.3.3 Blood Parameter Analysis

The blood samples were collected on week 7 from three groups via jugular venipuncture. The sample was analyzed using the MYTHIC 18 CBC machine to measure key haematological and biochemical parameters, including red and white blood cell counts, hemoglobin concentration, packed cell volume, and serum metabolites. These measurements provided indicators of the physiological condition and health status of the chickens under different dietary treatments.

3.3.4 Assessment of Carcass Quality

After 9 weeks, the animal from three group was slaughtered which later the carcasses were dissected manually and the following parameters were recorded: final live weight, kill-out weight, defeathered weight, carcass weight, dressing percentage, the weight of selected organs that include breast muscle, drumstick, wing, head, neck, shank, heart, liver, proventriculus, full gizzard and empty gizzard (Alghirani et al., 2021a; Chung et al., 2019).

3.3.5 Feed Cost Analysis (FCA)

Feed Cost Analysis (FCA) was calculated depending on the feeding cost/kg in live weight gain of crossbred village chicken. The cost includes the basic diet on commercial concentrate diet starter feed, commercial concentrate diet grower feed and dried BSFL as a feed supplement. The prices (RM/kg) are focused on:

a. Live crossbred village chicken
b. Commercial Concentrate Diet (Starter)
c. Commercial Concentrate Diet (Grower)
d. Dried Black Soldier Fly Larvae (BSFL)
e. Commercial Concentrate Diet (Starter) mix with 5% dried BSFL
f. Commercial Concentrate Diet (Grower) mix with 5% dried BSFL
g. Commercial Concentrate Diet (Starter) mix with 10% dried BSFL
h. Commercial Concentrate Diet (Grower) mix with 10% dried BSFL

3.4 Data Collection & Analysis

Data will be collected and recorded manually by tabulating using Microsoft Excel. The data analyses will use the IBM SPSS (Statistical Package for Social Science V.30) for parametric one-way ANOVA test and Tukey post hoc test to compare between the intervention group and the control group.

3.5 Ethical Consideration

The animal experiment will be conducted following the Ethical Committee for the Experimental Use of Animals at the Faculty of Veterinary Medicine, Universiti Malaysia Kelantan (UMK) under approval code UMK/FPV/ACUE/RES/002/2024.

CHAPTER 4

4.0 RESULTS

4.1 Nutritional Composition of Crossbred Village Chicken

Table 3 shows the nutrition composition of the feed based on the groups T1, T2, and T3. Based on the calories / energy (kcal/100g) shows that DBSFL is low in energy (219 kcal / 100g) compared with starter feed (346 kcal / 100g) and grower feed (370 kcal / 100g) under T1. However, when 5% and 10% of DBSFL was added into a commercial concentrate diet, a slight increase in calories / energy (kcal/100g) was shown in groups T2 and T3. Furthermore, when dried BSFL is added, crude fat and crude fiber is shown to increase slightly in groups T2 and T3 compared to T1 while slightly increasing in ash. Dried BSFL is low in moisture introduced in group T2 and T3 shows moderate low compared to T1.

TYPE		**T1**		**T2**		**T3**	
BIL	**PARAMETER**	**STARTER (S)**	**GROWER (G)**	**5% S + DBSFL**	**5% G + DBSFL**	**10% S + DBSFL**	**10% G + DBSFL**
1	Energy (kcal/100g)	346.00	370.00	357.00	374.00	362.00	373.00
2	Crude Protein (%)	23.90	19.30	23.20	20.60	23.50	21.00
3	Crude Fat (%)	3.50	5.50	4.80	6.30	6.30	7.00
4	Crude Fiber (%)	1.30	1.50	1.80	1.80	1.80	2.10
5	Ash (%)	5.60	4.40	5.40	4.80	5.70	5.90

6	Moister (%)	12.40	10.00	11.40	9.60	11.60	9.60

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet)

Table 3: Nutritional composition of the feeds, based on dry matter basis.

4.2 Blood Parameter Analysis

The study showed that dietary inclusion of dried BSFL significantly affected three blood parameters which are the packed cell volume (PCV), creatine kinase (CK) and Total Protein (TP). For PCV, the 10% DBSFL group (T3) displayed a significantly lower value compared to the 5% inclusion level (T2), while the control group (T1) remained statistically similarly to both.

In contrast, CK levels showed a significant increase in both DBSFL-supplemented groups (T2 and T3) compared to the control. The CK values in T2 and T3 shared the same superscript (“b”), meaning the two treatment levels did not differ from each other but are both significantly higher than T1 (“a”). Total Protein is higher in DBSFL-fed groups, indicating a clear improvement in protein status with DBSFL supplementation.

Overall, although significant differences is observed in PCV, CK, and Total Protein, all values remained within normal physiological ranges. These findings demonstrate that dried BSFL can be incorporated into the diet of crossbred village chickens without causing negative effects on blood health.

Table 4: Effects of DBSL on the blood parameter in Crossbred Village Chicken

Parameter	T1	T2	T3	*p*-Value	Standard Reference Range*
PCV (%)	38.67^{ab} ± 0.99	39.0^{a} ± 0.53	35.73^{b} ± 0.37	**0.03**	22 - 35
CK (U/L)	190.67^{a} ± 5.49	209.67^{b} ± 0.88	214.0^{b} ± 1.4	**0.01**	54 - 252
LDH (U/L)	177.4 ± 7.72	180.67 ± 2.33	184.53 ± 3.3	0.63	154 - 271
ALT (U/L)	15.73 ± 1.15	17.7 ± 0.26	18.53 ± 0.58	0.09	5 - 20
ALP (U/L)	175.0 ± 17.56	160.0 ± 0.58	196.1 ± 1.56	0.12	54 - 326
Total protein (g/dL)	2.1^{a} ± 0.06	2.53^{b} ± 0.03	2.57^{b} ± 0.12	**0.01**	2.0 - 3.0
Albumin (g/dL)	2.33 ± 0.15	2.6 ± 0.06	2.5 ± 0.15	0.14	0.75 - 4.90
Urea (mmol/L)	11.1 ± 3.6	12.93 ± 0.03	14.1 ± 0.26	0.62	0.6 - 124.0
Uric acid (mmol/L)	1.0^{a} ± 0	1.0^{a} ± 0	1.0^{a} ± 0	**0**	3.0 - 8.6
Triglyceride (mmol/L)	130.0 ± 5.13	128.63 ± 0.68	128.33 ± 2.4	0.93	109 - 271
Cholesterol (mmol/L)	202.33 ± 7.22	191.33 ± 1.86	187.1 ± 1.46	0.11	172 - 286

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet), *Reference range for avian from IDEXX VetTest Chemistry Analyzer, IDEXX Laboratories Inc.

Means$^{a, b, c}$ ± SE. Means with different superscripts in the same row differ significantly at ($p < 0.05$) with p values presented in bold,

4.3 Assessment of Carcass Quality

The final live weight, kill-out weight and de-feathered weight of all the carcasses from all the groups show no significant different however, the T2 group recorded the highest mean values. Only a few carcass components showed a significant difference ($p < 0.05$) which are the head weight, thigh weight, and heart weight. A significant increase in head weight is observed in T2 (5% DBSFL) compared to both T1 and T3. However, there is an absence of a similar increase in T3 (10% DBSFL).

For the edible part, the thigh of the control group shows a significant higher compared to the T2 and T3. Although the heart weight parameter showed a p-value of 0.04, all the treatment group shared the same transcript indicating that there are no significant pairwise differences among the group. This suggests that DBSFL inclusion at either 5% or 10% did not significantly influence the heart weight of the crossbred village chicken.

Table 5: Effects of DBSL on the carcass quality in Crossbred Village Chicken

Parameter	T1	T2	T3	*p*-Value
Final body weight (g)	1707.14 ± 69.38	1806.25 ± 55.45	1627.78 ± 77.33	0.2
Kill-out weight (g)	1527.14 ± 71.77	1650 ± 57.07	1492.22 ± 78.28	0.27
De-feathered weight (g)	1352.86 ± 79.63	1416.25 ± 46.94	1283.33 ± 44.28	0.25
Carcass weight (g)	1147.43 ± 52.133	1217.75 ± 35.04	1083 ± 47.49	0.12

Head (g)	50.57^{a} ± 4.29	72.25^{b} ± 1.46	57.11^{a} ± 4.19	**0.01**
Neck (g)	59.0 ± 11.07	81.25 ± 9.15	83.33 ± 8.33	0.17
Shank (g)	40.86 ± 3.60	43.88 ± 1.08	42.89 ± 3.09	0.76
Drumstick (g)	95.0 ± 5.0	100 ± 0	105.56 ± 5.56	0.26
Thigh (g)	127.14^{a} ± 10.85	106.25^{ab} ± 6.25	94.44^{b} ± 5.56	**0.02**
Wing (g)	96.43 ± 3.57	100 ± 0	100 ± 0	0.31
Breast (g)	257.14 ± 20.20	265.5 ± 20.59	288.89 ± 18.21	0.47
Spleen (g)	5.57 ± 0.75	6.38 ± 0.60	5.33 ± 1.0	0.65
Liver (g)	37.86 ± 2.27	40.25 ± 3.59	36.89 ± 1.76	0.64
Heart (g)	8.0^{a} ± 0.31	9.75^{a} ± 0.49	8.0^{a} ± 0.62	**0.04**
Gastrointestinal (g)	136.0 ± 5.67	127.63 ± 9.60	127.56 ± 5.71	0.67
Full gizzard (g)	38.14 ± 5.30	40.38 ± 1.70	33.22 ± 3.23	0.34
Empty gizzard (g)	28.0 ± 2.09	32.25 ± 1.29	27.11± 1.60	0.83

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet)

Means$^{a, b, c}$ ± SE. Means with different superscripts in the same row differ significantly at ($p < 0.05$) with p values presented in bold,

4.4 Assessment of Growth Performance

The growth performance assessment for the crossbreed villages chicken is highlighted in table 6. Statistically, there was no significant difference ($p>0.05$) in body weight gain and average weekly gain parameter. However, the feed conversion ratio shows statistically significant ($p<0.05$) between groups T1, T2, and T3.

Table 6: Total feed intake in this study, initial BW, final BW, BWG, average weekly gain, feed intake (DM kg/day), total feed intake per day (kg/day) and FCR.

		T1	T2	T3
TYPE	**BSFL**	-	5%	10%
	COMMERCIAL DIET	100%	95%	90%

BIL	PARAMETER					SEM	p-Value
1	Total feed intake	(g)	9419.00	8071.00	7843.00	-	<0.001
2	Initial body weight	(g)	41.70	42.80	44.30	1.80	0.167
3	Final body weight	(g)	1577.80	1565.00	1725.00	205.28	0.250
4	Body weight gain	(g)	6823.78	6860.40	7121.00	204.86	0.292
5	Average weekly gain	(g)	1113.30	1307.20	1225.00	13.11	0.333
6	Feed intake	(DM kg/day)	74.75	64.05	62.25	-	<0.001
7	Total feed intake per day	(kg/day)	149.50	128.11	124.50	-	<0.001
8	FCR		1.32	1.12	1.10	0.03	<0.001

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet), SEM = Standard error of mean.

4.5 Cost Analysis of Crossbred Village Chicken

In Table 7, the cost analysis (RM) of crossbreeding villages chicken in different groups shows that the fixed cost remains constant. However, the variable cost highlights that the cost of starter feed slightly reduced when dried BSFL was introduced in groups T2 and T3 similar to grower feed. This reduction was offset by the additional cost of incorporating dried BSFL into the feed, leading to a higher total feed cost in T2 and T3 compared to the control group T1.

Table 7: Cost analysis (RM) of crossbreed village chicken in different diets.

No	Cost Item	Unit	Price		
			T1	**T2**	**T3**
	A. Fixed Cost				
1	Chicken	chick	35.00	35.00	35.00
	B. Variable Cost				
1	201p Starter	kg	10.50	10.20	9.90
2	202p Grower	kg	235.40	212.00	194.30
3	Dried BSFL	kg	-	30.30	60.20
	Total Variable Cost (RM)		**245.9**	**252.50**	**264.40**

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet), Starter 201p = RM 2.90/kg, Grower 202p = RM 2.90/kg, Dried BSFL =RM 7.50/kg, Crossbred Villages Chicken = RM 3.50/chick

In table 8, the net profit is calculated by comparing the income generated by the chickens with the total production costs. For E (Net profit from live weight gain), the income from live weight gain (A) was subtracted by the sum of the daily feed cost (B) and fixed cost (C). This shows how much profit was obtained specifically from the weight gained during the 63-day feeding period.

Table 8: Cost analysis (RM) of crossbreed village chicken for feeding trial 63 days

	Diet		
	T1	**T2**	**T3**
A. Income from live weight gain (RM/day/ chicken)	20.83	18.74	22.92
B. Cost of feeding in 63 days (RM/day/ chicken)			
Starter Feed	0.10	0.10	0.10
Grower Feed	0.60	0.20	0.50
Dried BSFL		0.05	0.10
Total feed cost (RM/day/chicken)	0.70	0.35	0.70
C. Fixed cost in 63 days			
Chicks	3.50	3.50	3.50
D. Gross return over feed cost (RM/day/ chicken)	20.13	18.39	22.22
E. Net profit from live weight gain (RM/day/ chicken)	16.63	14.89	18.72

Note: **T1** = (Control), **T2** = (5% DBSFL, 95% Commercial Concentrate Diet), **T3** = (10% DBSFL, 90% Commercial Concentrate Diet), Starter 201p = RM 2.90/kg, Grower 202p = RM 2.90/kg, Dried BSFL =RM 7.50/kg, Live Crossbred Villages Chicken = RM 16/kg

CHAPTER 5

5.0 DISCUSSION

5.1 Nutritional Composition of Crossbred Village Chicken

To determine the nutritional composition of crossbred village chicken, the feed sample is evaluated using proximate analysis. In a study by Cherian (2020), he mentions that the nutrient analysis of the feed is done as a system to evaluate the feed and what is required by animals to enable producers and researchers to optimize nutrient utilization, which helps to address underperformance and lower costs of food production.

When introducing the dried BSFL in a commercial concentrate diet, the nutritional composition is different in terms of calories / energy, crude protein, crude fat, crude fiber, ash and moisture content compared to T1 (control group). Dried BSFL contains a high quantity of fat as energy for the development of pupation and is responsible for the high-fat content in diet or meal according to Oluokum (2000). However, with different growth stages, the crude fat of BSFL with the age of day 4 to day 14 increases drastically up to 28.4% while the crude protein decreases rapidly simultaneously with the development stage, Liu et al. (2017) mentions in a study that in adulthood, the maximum crude protein present is 57.6% and crude fat is 21.6%.

Based on a study by Afolabi (2021), physical and chemical properties of feed can be affected by the moisture levels in terms of the freshness and stability for long term storage in commercial concentrate diets. The dried BSFL, when added into commercial

concentrate diets in poultry diets displays low moisture content which then lowers the susceptibility towards microbes to prevent deterioration during storage (Afolabi et al., 2021).

Dried BSFL was discovered to be a good source of minerals and vitamins in a crossbred village chicken diet. Seyedalmoosavi (2022) mentions in a study BSFL contains a high amount of calcium that can reach up to 9% of dry matter which is considered a main advantage. In contrast, Afolabi (2021) studies that excessive ash in poultry diets causes formation of crystals in the urinary tract, bone and causes joint problems at the grower stage. Thus, a high content in ash had never been proven beneficial towards broilers and layers in previous literature (Afolabi et al., 2021).

Mwaniki et al (2018) states that applying an insect-based diet in birds increases the fiber content in different growing stages of BSFL, which in turn increases the feed intake. The fibers help in increasing ceca fermentation in birds, resulting in better nutrient absorption and development (Bovera et al., 2016). Feeding fiber in lower rations is preferred to show positive effect, however poor bird performance is proven to have an impact on voluntary feed intake significantly and nutrient digestibility when more than 3% in ration is given (Tejeda et al., 2021).

5.2 Blood Parameter Analysis

Packed cell volume can be defined as the percentage of erythrocytes in the blood compared to the serum component (Calhoun, 2008). It would help diagnose anemia and the hydration status of the broilers that guide the course of treatments. The PCV value of

all the group was a bit higher from the normal range as the normal range were found between 22-35% according to Odunitan-Wayas et al., 2018. Although in the current study, PCV was slightly lower in the 10% DBSFL group (T3) compared to 5% DBSFL group (T2) while control group (T1) is slightly higher compared to group (T2). The increasement of the PCV could be affected by the hydration status of the crossbred village chicken. This prove where the water restriction results in hemoconcentration reflected by increases in packed cell volume (Iheukwumere and Herbert, 2003; Mushi et al., 1999) indicating no adverse effect on overall red blood cell volume or oxygen-carrying capacity.

Serum biochemistry evaluation in feed additives study offers valuable information on nutrient metabolism and possible body alterations caused by intrinsic and extrinsic factors (Basit et al., 2020c). Hence, several biochemical parameters are evaluated in this study that were harvested from the crossbreed village chicken serum. Elevated serum ALT, AST, CK and LDH levels in broilers are associated with muscle or liver damage (Kong et al., 2021). Any damage in muscle tissue could compromise the integrity of the sarcolemma, which leads to the leaking of the creatine kinase (CK) and lactate dehydrogenase (LDH) enzymes into the serum (Kong et al., 2021). The current study reported serum CK levels increased in both DBSFL groups (T2 and T3) compared to the control. The observed increase in serum creatine kinase (CK) levels in crossbreed village chicken may be attributed to an increase in skeletal muscular injury caused by the feed additives, regardless of the significant differences, all groups in this experiment had a normal serum creatine kinase (CK) level.

Serum proteins, synthesized in the liver, serve multiple functions, including the maintenance of blood volume through colloidal osmotic pressure, acting as blood pH buffers, transporting enzymes, drugs and hormones, and participating in processes such as cell coagulation, metabolism and the immune response against antigens (Rezende et al., 2017). There is a significant increase in serum total protein in both T2 and T3 groups compared to the control group. Increase of the total protein could be influenced also by the hydration status where the water restriction also will result in hemoconcentration reflected by elevated levels of total proteins (TPs), albumin (ALB) and globulin (GLOB; Cork and Halliwell, 2002).

5.3 Assessment of Carcass Quality

Ideal broilers should possess high slaughter yields, desirable carcass conformation scores, proper aesthetic, nutritional and healthy characteristics (Davoodi & Ehsani, 2020). In this study, the final live weight, kill-out weight, de-feathered weight and carcass weight from the T2 groups were consistently higher to the T3 group and control group despite not being statistically significant. The enhanced carcass weight observed could be because BSFL are proven to be rich in essential amino acids, such as lysine and methionine, which are important for poultry production (Spranghers et al., 2017).

In this current study, only a few carcass components showed a significant difference ($p < 0.05$) which are the head weight, thigh weight, and heart weight. Overall, the significant differences observed were limited to the certain organ and not the major carcass component such as the final body weight, carcass weight or breast. This is proven

by Pieterse et al. (2019) where meat quality of chickens fed with BSF has been similar to that of the chickens fed commercial diet, therefore, they can be used as a partial substitution for soybean meals.

5.4 Assessment of Growth Performance

In commercial concentrate diets, the impact of dried BSFL shows positive results and is observed in other research studies in poultry to increase body weight, body weight gain and feed conversion ratio (FCR) (De Souza Viela Jessica et al., 2021). The feed efficiency which is measured by the feed conversion ratio (FCR), with lower values indicating higher efficiency stated by Huang et al (2022). In this experiment, the body weight gains are higher and lower FCR values were observed consistently in the experimental groups compared to the control group.

Huang et al (2021) mentions that the feed conversion ratio (FCR) measures feed efficiency, with lower values indicating higher efficiency. In this study, higher body weight gains and lower FCR values are observed consistently in the experimental groups compared to the control group. This might be possible that the chitin content of the BSFL (4.62% DM) acting as a prebiotic in the intestine of broilers as described, resulting in decreased DM digestibility and improved FCR (Bovera et al., 2016).

Studies by Dabbou et al (2018) on Ross 308 chickens, that minimal changes in feed intake, feed conversion ratio (FCR) and body weight gain are observed at lower

BSFL levels (5% to 10%). However, there was a significant reduction in BWG noticed when BSFL is given at the highest level of 15%.

5.5 Cost Analysis of Crossbred Village Chicken

The findings of this study contrast with those reported by Onsongo et al (2018) that the cost of feed consumed by the birds reduces with increasing levels of BSFL meal as a replacement for conventional fish-meal sources in broiler diets. However, Chia et al (2019) reports that there is improved economic performance with increased integration of higher inclusion levels of BSFL meals, which demonstrates the sustainability and viability of BSFL meal as a promising alternative protein source to close the nutrient gap in the animal feed industry. Ultimately, the economic viability of incorporating BSFL into poultry diet.

Overall, the inclusion of 10% dried BSFL in the diet of crossbred village chickens improved profitability without increasing daily feed cost compared with the control. Although 5% BSFL reduced feed cost, it did not translate into improved income or net profit. These findings suggest that 10% BSFL inclusion is a more economically beneficial feeding strategy, likely due to enhanced growth performance and feed efficiency.

CHAPTER 6

6.0 CONCLUSION AND RECOMMENDATION

6.1 Conclusion

In conclusion, the use of dried BSFL at 5% and 10% in the diets of crossbred village chickens has been shown not to adversely affect physiological health and carcass quality of the chicken. This suggests that BSFL does not cause physiological stress or metabolic disturbances, further supporting its suitability as a stable and healthy dietary ingredient.

The incorporation of dried BSFL (dried Black Soldier Fly Larvae) as an alternative protein source in poultry diets, has shown positive effects on the growth performance of crossbred village chickens. Current findings indicate that inclusion levels of 5% and 10% dried BSFL do not negatively affect growth performance, demonstrating that these levels are both safe and nutritionally appropriate for use in poultry rations.

Moreover, dried BSFL has the potential to improve carcass quality, likely due to its high nutrient density, beneficial amino acid profile, and bioactive components. Enhancements in carcass traits may include better muscle development, improved dressing percentage, or overall improved meat quality, depending on the level of inclusion.

While the cost of incorporating dried BSFL may be slightly higher than using commercial concentrate diets alone, the additional expense varies depending on the

farmer's objectives, feed formulation approach, and availability of BSFL. Despite the potential increase in cost, dried BSFL remains a sustainable and highly promising alternative protein source.

Therefore, dried BSFL is recommended as a viable substitute for conventional protein ingredients in the diets of crossbred village chickens, offering benefits in growth performance, physiological stability, and potential improvements in carcass quality.

6.2 Recommendation for Future Research

Future research should consider increasing the sample size to at least 30 animals per experimental group, as a larger number of subjects can improve the statistical power of the study and help produce more reliable and meaningful differences in growth performance. Additionally, the analysis of dietary samples should be expanded by including a greater variety of nutritional components and ensuring that each sample is tested in duplicate or triplicate. This will enhance the accuracy and consistency of the proximate analysis results.

Moreover, the study should evaluate a wider range of inclusion levels for dried BSFL as an alternative feed ingredient. For instance, incorporating higher percentages such as 5% and 15% dried BSFL would allow researchers to better understand the dose-dependent effects on growth performance, nutrient utilization, and overall animal health. Exploring these broader inclusion rates would also help identify the optimal level of dried BSFL that can be used effectively in practical feeding programs.

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APPENDIX A

A1: Preparation cage for grower stage.

A2: Setup the automatic drinker.

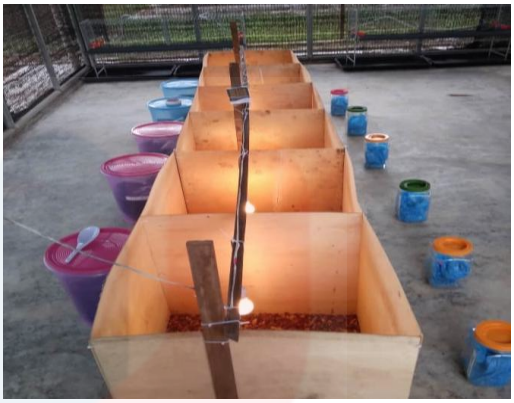
A3: Preparation feed for each group.

A4: Disinfect the area using Lindores.

A5: Foot dip at the front door.

A6: receiving DOC from supplier.

A7: Chicken at starter feed stage.



A8: Brooding area for DOC.

A9: Identification of chick by tagging.

A10: Area for grower chicken.

A11: Deep cleaning after finishing.

A12: Air dried the equipment.

A13: Halal slaughtering for harvesting.

A14: Collection of blood via jugular vein.

A15: De-feathering weight was taken.

A16: Head and shank of chicken.

A17: Cutting chicken for carcass quality assessment.

A18: Internal organ of chicken.

APPENDIX B

Treatment	Week 1	Week 6	Week 9
T1	 Day 7	 Day 42	 Day 63
T2	 Day 7	 Day 42	 Day 63
T3	 Day 7	 Day 42	 Day 63

B1: Growth performance of the crossbred villages chicken