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# Comparative evaluation of *Acheta domesticus* and *Hermetia illucens* as alternative protein sources for the growth, health, and meat quality of the broiler

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**Introduction:** Insects are receiving increasing attention due to their potential to enhance farming efficiency and sustainability, mitigate vast quantities of bio-waste, and improve animal performance. It is hypothesized that replacing soybean meal with *Acheta domesticus* will improve broiler productive performance, health, and meat quality. Therefore, the objective of this study was to assess the effect of the gradual replacement of soybean meal (4%, 8%, and 12%) with house cricket (*A. domesticus*) and black soldier fly (*Hermetia illucens*) on productive performances, hematology, intestinal morphology, and meat quality attributes of male broiler (Ross 308).

**Methods:** A total of 350 1-day-old chicks ( $39.23 \pm 0.19$  g) were divided into seven groups (five pens per group and 10 chicks per pen) following a completely randomized design. The seven groups included 4%, 8%, and 12% SBM replacements with *A. domesticus* and *H. illucens*. Soybean meal was the basal diet considered the control.

**Results:** The broilers fed 12% *A. domesticus*, or 12% *H. illucens* had significantly higher ( $p < 0.05$ ) live weight, average daily weight gain, and improved feed conversion ratio than the broilers fed basal (SBM) diet throughout starter, grower, and finisher phases. Hematology (complete blood count and serum biochemistry traits) and intestinal morphology (villus height, villus width, crypt depth, and villus height-to-crypt depth ratio) of the broilers improved when fed on 12% *A. domesticus* and 12% *H. illucens* meals as compared to the control group ( $p < 0.05$ ). Feeding 12% *A. domesticus* and 12% *H. illucens* also had a significant positive impact on the meat quality traits of broilers, such as maximum redness (a\*) and yellowness (b\*) with minimal cooking loss and lightness (L\*) ( $p < 0.05$ ) than the broilers fed the control diet.

**Discussion:** Using insect meal as an alternative to traditional feed sources could contribute significantly to the sustainable expansion of the poultry industry.

#### KEYWORDS

*Acheta domesticus*, broiler, hematology, *Hermetia illucens*, productive performance, replacement

## 1 Introduction

Soybean meal (SBM) and fish meal (FM) are the predominant protein sources in poultry feed. Still, there is limited land available worldwide for soybean cultivation. Overexploitation of marine resources has led to a significant decline in small pelagic forage fish, which are critical for producing FM and fish oil (Veldkamp et al., 2022). Moreover, the increasing expenses associated with these traditional protein sources posed a threat to the sustainable future of the poultry industry (McMichael and Bambrick, 2005). Furthermore, food insecurity is a pressing issue in numerous developing countries, and there are expected challenges in supplying food for a projected population of over 9 billion people in 2050 (Makkar et al., 2014). This situation has intensified the quest for alternative protein sources to fulfill the nutritional demands of humans and animals.

Insects are emerging as a novel alternative protein source, promoting sustainable growth in the poultry sector (Sánchez-Muros et al., 2014). They have the potential as a standard protein source in animal feedings, i.e., poultry, pigs, and fish, owing to their high-quality protein content (25%–75% on a DM basis) and suitable amino acid composition and fatty acid composition (n-3 FA) (Henry et al., 2015; Kulma et al., 2020; Van Huis, 2013; Veldkamp and Bosch, 2015). Moreover, they have minimal competition with human food resources and are eco-friendly in terms of energy cost, land, and footprints (Ballitoc and Sun, 2013), which makes insects a promising option from an ecological standpoint (Makkar et al., 2014; Oonincx and De Boer, 2012; Sánchez-Muros et al., 2014).

Several types of insects, including black soldier fly (*Hermetia illucens* L.), yellow mealworm (*Tenebrio molitor* L.), house fly (*Musca domestica* L.), mulberry silkworm (*Bombyx mori* L.), and grasshopper (*Caelifera* spp.), have been recognized as promising sources of protein to replace conventional protein sources in animal feeds (Van Huis et al., 2013). Free-range birds voluntarily consume insects as a natural protein source (Adetunmbi, 2023; Hwangbo et al., 2009; Ijaiya and Eko, 2009; Khatun et al., 2003; Sajjad et al., 2024a). The nutritional value of insects can oscillate with the species, rearing substrate, development stages, and handling methods (Danieli et al., 2019; Gasco et al., 2020; Slimen et al., 2023). Incorporating insects into poultry feed instead of SBM or FM did not adversely affect the broilers' growth performances (Adeniji, 2007; Oyegoke et al., 2006; Wang et al., 2005). Moreover, insect meal improves carcass traits in terms of dressing percentage, breast muscle, thigh muscle, slaughter, dressed

carcass, and eviscerated weights in broilers (Ballitoc and Sun, 2013; Hwangbo et al., 2009; Khatun et al., 2003; Sajjad et al., 2024b).

Previous studies have demonstrated that the growth performance, blood hematology, gut morphology, and meat quality of broilers (Benzertiha et al., 2019), Japanese quails (Zadeh et al., 2019), and barbary partridges (Loponte et al., 2017) are improved by the inclusion of *H. illucens* and *T. molitor* in their diets (Benzertiha et al., 2020; Khan et al., 2018). The growth performance and nitrogen balance of Ross 308 are enhanced with 2.6% *H. illucens* meal in the starter phase (Neumann et al., 2018). Moreover, meals containing 5% to 7.5% *H. illucens* improved the feed efficiency and increased thigh weight while reducing meat pH, whereas 10% *H. illucens* boosted growth in Cobb 500 broiler chickens (Dahiru et al., 2016). Furthermore, it was noted that substituting partial or all of the soybean oil with *H. illucens* larvae fat had no adverse impact on the growth performance of young turkeys (Sypniewski et al., 2020). The inclusion of 20% *H. illucens* in the diet of Ross 308 male broilers improved meat quality by elevating levels of lauric acid, myristic acid, and eicosapentaenoic fatty acid, while also slightly decreasing the total polyunsaturated fatty acid (PUFA) content (Vilela et al., 2021). The diet with 5% *H. illucens* resulted in a decrease in abdominal fat; 10% *H. illucens* led to an increase in carcass and breast weight, while 15% inclusion of *H. illucens* increased the body weight, abdominal fat, meat redness, protein content in meat, and the levels of monounsaturated fatty acids (MUFAs) in breast meat while reducing PUFA in breast meat (Schiafone et al., 2019). Gut health and functioning are mainly associated with the intestinal morphology of the broilers, which is affected by the dietary protein level and digestibility of the ingested food (Kuzmuk et al., 2005; Laudadio et al., 2012; Qaisrani et al., 2014). Intestinal morphology can be assessed with the development of morphometric indices such as villus height (Vh) and crypt depth (Cd) (Franco et al., 2006; Laudadio et al., 2012).

The poultry industry depends on SBM to meet the demand for protein in animal feeds, as only a few insect species are currently utilized at an industrial scale (Sajjad et al., 2023). Therefore, it is a smart choice to focus on local insect species as substantial feed ingredients in animal sectors (Sajjad et al., 2024c).

The house cricket (*Acheta domesticus* L.) (Orthoptera: Gryllidae) has been utilized globally as feed for insectivores (Kulma et al., 2019) because of its high protein content (60%–70% dry weight) with all essential amino acids and 10%–23% lipids (Udomsil et al., 2019). Moreover, it has high levels of omega-3 and omega-6 fatty acids and minerals such as P, Na, and Ca (Han and Heinonen, 2022; Oonincx

and Finke, 2021). Furthermore, it can potentially transform organic wastes, including food scrap and agricultural by-products, into nutritious biomass that is beneficial for animal feeds, particularly for poultry and aquaculture (Van Peer et al., 2021). Cricket farming generates high-quality by-products, which can be used as organic fertilizer (Halloran et al., 2017). Furthermore, the European Union has recently authorized the inclusion of *H. illucens*, *M. domestica*, *T. molitor*, *Alphitobius diaperinus*, *A. domesticus*, *Gryllobates sigillatus*, and *Gryllus assimilis* as a protein source in poultry and pig feed (Schiavone and Castillo, 2024); however, there is limited information regarding the effects of partial replacement of SBM with *A. domesticus* on the productive performance, health, and meat quality traits of Ross 308 broiler. It is hypothesized that the replacement of SBM with *A. domesticus* will improve broiler productive performance, health, and meat quality. The aim of the present study was to evaluate the effects of replacing 4%, 8%, and 12% SBM with *A. domesticus* and *H. illucens* on the productive performance, hematology, intestinal morphology, and meat quality of broiler chickens.

## 2 Materials and methods

### 2.1 Institutional review board

The biological trials on broiler Ross 308 were carried out for 35 days under controlled conditions in A block, University of Animal and Veterinary Sciences (UVAS), Ravi Campus, Pattoki, Lahore, Pakistan. The Ethical Review Committee (No. DR/495) at UVAS approved all the procedures.

### 2.2 Insects

Initially, house crickets (*A. domesticus*) were collected using an aerial net from neglected kitchens of homes and hotels, and then the stock culture on chicken waste was established, while black soldier flies *H. illucens* were also obtained from the established culture under controlled conditions (28°C ± 2°C, relative humidity 65% ± 5%, and 16 h light:8 h dark) at the Department of Entomology, the Islamia University of Bahawalpur. Mature larvae of *H. illucens* (18 days old) and adult *A. domesticus* (70 days old) were harvested from the chicken waste and boiled at 100°C for 3 min to ensure they were killed, and feed and fecal remnants were removed and subsequently oven-dried at 60°C for 24 h (Tan et al., 2018). The dried insects were ground in a blender and stored at -20°C. The larvae of *A. domesticus* and *H. illucens* were analyzed for their nutritional compositions, energy contents, and amino acids (% on DM basis) using proximate analyses and an amino acid analyzer at the Department of Animal Nutrition at UVAS (Table 1).

### 2.3 Experimentation

Male Ross 308 was used to assess the impacts of 4%, 8%, and 12% substitution of SBM with *A. domesticus* and *H. illucens*. A total of 350 1-day-old chicks (39.23 ± 0.19 g) were allotted to seven dietary treatment

TABLE 1 The nutrients, energy contents, and amino acids of *H. illucens* and *A. domesticus* larvae.

Nutrients (%) <sup>a</sup>	<i>H. illucens</i>	<i>A. domesticus</i>
Dry matter	90.34	92.00
Crude protein	41.23	49.00
Ether extracts	31.00	11.20
Ash	11.00	5.60
Crude fiber	9.70	9.34
Nitrogen free extract (NFE)	5.50	22.00
Calcium	2.15	0.90
Phosphorus available	0.90	0.45
Energy levels (kcal/kg)	<i>H. illucens</i>	<i>A. domesticus</i>
Gross energy	4,948	4,320
Metabolizable energy <sup>b</sup>	1,474	1,294
Essential amino acid (%)	<i>H. illucens</i>	<i>A. domesticus</i>
Arginine	2.31	4.82
Lysine	3.23	6.45
Methionine	1.28	2.27
Threonine	1.82	3.38
Leucine	3.08	6.82
Isoleucine	2.64	3.75
Valine	3.18	4.16
Dispensable amino acid (%)	<i>H. illucens</i>	<i>A. domesticus</i>
Cysteine	0.43	3.40
Tryptophan	0.30	1.42
Glycine	3.10	3.20
Glutamic acid	11.20	11.00
Proline	2.92	2.27
Tyrosine	3.27	3.08
Phenylalanine	4.27	3.10

<sup>a</sup>All the analyses of each sample were replicated three times.

<sup>b</sup>Metabolizable energy (M.E.) was estimated by following Ravindran et al. (2014).

groups, with five pens per treatment and 10 birds per pen. The birds were placed in spotless and disinfected pens equipped with feeders, drinkers, heat lamps, and bedding materials (3 to 4 inches of rice husk). The chicks had *ad libitum* feed, water, and light. Temperature was maintained at 35°C during the initial week, subsequently adjusting to 28 ± 2°C till the completion of experiments, while the relative humidity was kept at 50% ± 5%. The photoperiod was maintained at 8 h of light and 6 h of darkness using artificial lighting. The birds were vaccinated for numerous diseases as per the set protocols by the veterinarian. Enclosures were inspected daily to assess clinical signs and record any mortality among the birds.

## 2.4 Formulation

SBM was progressively substituted at the levels of 4%, 8%, and 12% with *A. domesticus* and *H. illucens* by following the guidelines at the levels of Ross (2022). All the dietary treatments were formulated into three different phases, i.e., starter (1–10 d), grower (11–24 d), and finisher (25–35 d). Ingredients, along with the calculated and analyzed profiles of the starter, grower, and finisher meals, are presented in Tables 2–4.

## 2.5 Productive performances

Bird's performances, such as live weight (LW), average daily weight gain (ADG), daily feed intake (DFI), and feed conversion ratio (FCR), were calculated over a 35-day feeding period. The LW was recorded individually at the onset of the experiment and subsequently at intervals of 10 d, 24 d, and 35 d. Daily weight gain and feed intake were determined on both individual and pen bases following each growth phase, whereas the feed conversion ratio was calculated for each growth phase and for the intact intervals of the trial.

## 2.6 Hematology

On day 35, two chicks per pen were euthanized shortly after their arrival by a skilled professional using the rapid decapitation technique. Consequently, 2.5 mL of blood was collected into EDTA and serum-separating tubes. Hematology and serum biochemistry parameters were tested by following Campbell (1995).

## 2.7 Intestinal morphology

The small tissues (2–3 mm) from jejunum (JE) and ileum (IL) sites were excised from the slain birds, promptly washed with normal saline, and stored for 72 h in 10% formalin solution by following Laudadio et al. (2012). The transverse sections were studied by taking images under a light microscope, and villus height (Vh), villus width (Vw), crypt depth (Cd), and Vh/Cd were determined by the ImageJ software (Ferreira and Rasband, 2012).

## 2.8 Meat quality

Pectoralis major was used to evaluate meat quality parameters such as meat pH, drip loss, cooking loss, shear force, and meat color, including lightness (L\*), redness (a\*), yellowness (b\*), chroma (C\*), and hue (H\*) from the broiler. The deboned meat samples were refrigerated at 4°C overnight before the analyses. Cooking and drip losses were assessed as described by Kaić et al. (2021) and Zaid et al. (2020). Meat pH was tested using pH meter 3210 SET 2 from breast samples. The CR-410 colorimeter was used to determine meat color by following Priolo et al. (2002). The breast meat was cut into a rectangular shape (1 H × 1 W × 2 L, in cm) parallel to muscle fibers using a scalpel handle blade. TAXT plus 100C texture analyzer was used to assess the Warner–Bratzler shear force (N/cm<sup>2</sup>) with the

help of a V-slot blade. These analyses were carried out from UVAS, Lahore.

## 2.9 Statistical analysis

The pens served as the trial units for assessing productive performances, while individual birds were used to determine hematology, intestinal morphology, and meat characteristics. The significant differences among the treatment groups were analyzed using a completely randomized design (CRD) with a one-way analysis of variance followed by the Duncan Multiple Range (DMR). Significant statistical differences were accepted at  $p < 0.05$ , while values in the range of  $0.05 \leq p < 0.10$  were considered to indicate a trend. Orthogonal polynomial contrasts were used to assess the linear and quadratic effects. A general linear model (GLM) was performed to assess the impacts of dietary groups, intestinal sections, and their interactions. All the analyses were performed with IBM SPSS Statistics software (version 21 for Windows, SPSS Inc., Chicago, IL, USA).

## 3 Results

### 3.1 Productive performances

The productive performances, i.e., live weight, average daily weight gain, daily feed intake, and feed conversion ratio of the broilers fed with different meals, are summarized in Table 5. These parameters significantly differ ( $p < 0.05$ ) among all the dietary treatments except for the daily feed intake ( $p > 0.05$ ) across the feeding intervals. In starter and finisher phases, the LW was the highest in 12% *A. domesticus* and 12% *H. illucens* and the lowest in the control treatment. It was the maximum in 12% *A. domesticus* and 12% *H. illucens* and the minimum in 4% *H. illucens* in the grower phase. The LW was affected in a linear and quadratic manner among the *H. illucens* meals during the starter and finisher phases, while a linear response was seen during the grower phase. Similarly, LW responded linearly and quadratically to the *A. domesticus* meals during the feeding intervals.

During days 1–10, daily weight gain was the maximum in 12% *A. domesticus* and the minimum in the control group. During days 11–24, it was the highest in 12% *A. domesticus* and the lowest in 4% *H. illucens*. Similarly, maximum ADG was found in meals containing 12% *A. domesticus* and 12% *H. illucens* on days 25–35, and the minimum was found in the remaining treatments, whereas the highest daily weight gain was recorded in 12% *H. illucens* and the lowest in the control group. Polynomial contrast presented linear and quadratic effects in ADG during days 1–10, while a linear response was seen during the remaining feeding periods among the meals of *H. illucens*; likewise, there were linear and quadratic effects in ADG across the feeding periods among the *A. domesticus* meals.

The maximum FCR was registered in the broilers fed the control meal, while the minimum was registered when fed the 12% *A. domesticus* and 12% *H. illucens* across the feeding intervals.

TABLE 2 The ingredients, chemical compositions, and energy contents of starter meals.

Ingredients (%)	Starter meal						
	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Corn grain	51.17	51.93	54.35	55.57	52.67	54.40	55.28
Wheat bran	4.00	4.00	3.00	3.00	4.00	3.28	3.20
Rice polishing	4.00	4.00	4.00	4.00	4.00	4.00	3.75
Soybean oil	4.00	3.00	1.80	0.80	3.00	2.50	2.40
Soybean meal <sup>a</sup>	28.50	25.00	21.00	17.00	24.07	19.65	15.2
Fish meal <sup>b</sup>	6.00	6.00	6.00	6.00	6.00	6.00	6.00
HI and AD meals	–	4.00	8.00	12.00	4.00	8.00	12.00
L-Lysine HCl	0.03	–	–	–	–	–	–
DL-Methionine	0.15	0.12	0.10	0.07	0.10	0.04	0.07
Common salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Limestone	1.75	1.55	1.35	1.16	1.34	1.32	1.30
Vitamin premix <sup>c</sup>	0.05	0.05	0.05	0.05	0.47	0.46	0.45
Micro min premix <sup>d</sup>	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Total	100	100	100	100	100	100	100
Nutrients (%)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Dry matter	89.40	89.40	89.50	89.40	89.32	89.50	89.40
Crude protein	23.00	23.03	23.00	22.98	23.02	23.00	22.99
Ether extracts	6.65	6.72	6.80	6.86	6.56	6.56	6.61
Ash	4.06	4.22	4.27	4.36	4.24	4.29	4.36
Crude fiber	3.62	3.85	3.96	3.80	3.60	3.72	3.71
Nitrogen-free extract (NFE)	61.00	60.98	60.84	60.94	61.51	61.36	61.31
Calcium	0.95	0.95	0.95	0.95	0.95	0.95	0.95
Phosphorus available	0.51	0.51	0.52	0.52	0.51	0.52	0.52
Lysine	1.32	1.32	1.33	1.33	1.32	1.33	1.33
Methionine	0.55	0.55	0.55	0.55	0.55	0.55	0.55
Threonine	0.88	0.89	0.90	0.90	0.89	0.90	0.90
Valine	1.03	1.04	1.03	1.04	1.04	1.03	1.04
Arginine	1.41	1.41	1.42	1.40	1.41	1.42	1.40
Leucine	1.46	1.46	1.44	1.45	1.46	1.44	1.45
Isoleucine	0.88	0.89	0.88	0.89	0.89	0.88	0.89
Energy levels (kcal/kg)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Gross energy	4,606	4,593	4,598	4,604	4,596	4,598	4,604
Metabolizable energy <sup>e</sup>	2,980	2,978	2,973	2,974	2,978	2,973	2,973

<sup>a</sup>Crude protein contents of soybean meal was 45% according to (DM basis).

<sup>b</sup>Crude protein contents of soybean meal was 66% according to (DM basis). Cont., Control; HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*.

<sup>c</sup>Starter vitamin premix supplied per kg of diet: vitamin A IU: 12,000, vitamin D3 IU: 3,500, vitamin E: 30 mg, vitamin K: 3.0 mg, vitamin B1: 3.0 mg, vitamin B2: 8.0 mg, vitamin B6: 5.0 mg, vitamin B12: 0.020 mg, niacin: 40 mg, pantothenic acid: 18 mg, folic acid: 2.5 mg, biotin: 0.24 mg.

<sup>d</sup>Starter mineral premix: manganese: 120 mg, zinc: 100 mg, iron: 70 mg, copper: 8.0 mg, selenium: 0.240 mg, iodine: 1 mg.

<sup>e</sup>Metabolizable energy was calculated by following Ravindran et al. (2014) while all other ingredients had been analyzed. HI, *Hermetia illucens*; AD, *Acheta domesticus*.

TABLE 3 The ingredients, chemical compositions, and energy contents of grower meals (% as fed).

Ingredients (%)	Grower meal						
	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Corn grain	53.57	55.14	56.94	58.44	55.46	57.20	58.80
Wheat bran	5.00	5.00	4.00	3.00	5.00	4.00	3.25
Rice polishing	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Soybean oil	4.53	3.30	2.30	1.90	3.50	3.15	2.80
Soybean meal	26.00	22.00	18.30	14.50	21.20	16.90	12.47
Fish meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00
HI and AD meals	–	4.00	8.00	12.00	4.00	8.00	12.00
DL-Methionine	0.14	0.11	0.09	0.06	0.07	0.02	–
Common salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Limestone	1.36	1.05	0.97	0.70	1.05	1.03	1.01
Vitamin premix <sup>a</sup>	0.05	0.05	0.05	0.05	0.37	0.35	0.32
Micro min premix <sup>b</sup>	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Total	100	100	100	100	100	100	100
Nutrients (%)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Dry matter	89.40	89.30	89.50	89.40	89.30	89.50	89.40
Crude protein	21.51	21.50	21.52	21.50	21.50	21.52	21.50
Ether extract	7.06	7.08	7.10	7.14	7.00	7.00	6.99
Crude fiber	4.06	4.18	4.27	4.31	4.15	4.17	4.19
Ash	3.37	3.55	3.68	3.81	3.44	3.55	3.65
Nitrogen-free extract (NFE)	62.16	62.20	61.89	61.13	62.67	62.68	62.46
Calcium	0.75	0.75	0.75	0.75	0.75	0.75	0.75
Phosphorus available	0.43	0.42	0.42	0.43	0.42	0.42	0.43
Lysine	1.18	1.17	1.20	1.18	1.19	1.19	1.19
Methionine	0.50	0.52	0.52	0.49	0.51	0.51	0.51
Threonine	0.80	0.81	0.79	0.80	0.79	0.79	0.79
Valine	0.90	0.91	0.90	0.92	0.92	0.91	0.92
Arginine	1.28	1.27	1.29	1.28	1.27	1.28	1.27
Leucine	1.29	1.28	1.30	1.31	1.30	1.31	1.30
Isoleucine	0.80	0.82	0.81	0.82	0.81	0.80	0.80
Energy levels (kcal/kg)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Gross energy	4,663	4,659	4,664	4,671	4,659	4,643	4,645
Metabolizable energy <sup>c</sup>	3,055	3,049	3,048	3,051	3,049	3,048	3,051

Cont., Control; HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*.

<sup>a</sup>Grower vitamin premix supplied per kg of diet: vitamin A IU: 9,000, vitamin D3 IU: 3,000, vitamin E: 25 mg, vitamin K: 3.0 mg, vitamin B1: 2.4 mg, vitamin B2: 6.5 mg, vitamin B6: 4.2 mg, vitamin B12: 0.015 mg, niacin: 35 mg, pantothenic acid: 15 mg, folic acid: 1.5 mg, biotin: 0.21 mg.

<sup>b</sup>Grower minerals premix: manganese: 100 mg, zinc: 80 mg, iron: 60 mg, copper: 8.0 mg, selenium: 0.240 mg, iodine: 1 mg.

<sup>c</sup>Metabolizable energy was calculated by following Ravindran et al. (2014) while all other ingredients had been analyzed. HI, *Hermetia illucens*; AD, *Acheta domesticus*.

TABLE 4 The ingredients, chemical compositions, and energy contents of finisher meals (% as fed).

Ingredients (%)	Finisher meal						
	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Corn grain	57.22	58.60	60.94	63.18	59	61.06	62.90
Wheat bran	5.00	5.00	4.00	3.00	5.00	4.25	3.24
Rice polishing	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Soybean oil	5.52	4.33	3.23	2.03	4.80	4.10	3.70
Soybean meal	22.50	18.50	14.50	10.70	17.54	13.02	8.65
Fish meal	4.00	4.00	4.00	4.00	4.00	4.00	4.00
HI and AD meals	–	4.00	8.00	12.00	4.00	8.00	12.00
L-Lysine HCl	0.04	–	–	–	–	–	–
DL-Methionine	0.14	0.06	–	0.06	0.07	0.02	–
Common salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Limestone	1.15	1.15	1.15	0.60	0.91	0.90	0.88
Vitamin premix <sup>a</sup>	0.05	0.05	0.05	0.05	0.32	0.3	0.28
Micro min premix <sup>b</sup>	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Total	100	100	100	100	100	100	100
Nutrients (%)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Dry matter	89.45	89.20	89.40	89.50	89.30	89.60	89.50
Crude protein	19.52	19.57	19.48	19.52	19.52	19.72	19.53
Ether extract	8.06	8.10	8.13	8.13	8.02	8.02	8.02
Crude fiber	4.00	4.02	4.10	4.10	4.00	4.08	4.04
Ash	3.48	3.50	3.51	3.57	3.46	3.44	3.51
Nitrogen-free extract	63.80	63.70	63.55	63.40	64.26	64.24	64.15
Calcium	0.64	0.67	0.65	0.66	0.66	0.65	0.66
Phosphorus available	0.34	0.35	0.38	0.36	0.36	0.36	0.36
Lysine	1.10	1.08	1.07	1.08	1.08	1.09	1.08
Methionine	0.50	0.48	0.49	0.48	0.48	0.48	0.48
Threonine	0.70	0.71	0.74	0.72	0.72	0.72	0.72
Valine	0.80	0.82	0.85	0.85	0.84	0.84	0.85
Arginine	1.20	1.17	1.18	1.18	1.18	1.17	1.18
Leucine	1.18	1.20	1.19	1.20	1.19	1.19	1.20
Isoleucine	0.78	0.73	0.75	0.75	0.75	0.76	0.75
Energy levels (kcal/kg)	Cont.	HI4	HI8	HI12	AD4	AD8	AD12
Gross energy	4,738	4,733	4,723	4,713	4,723	4,721	4,716
Metabolizable energy <sup>c</sup>	3,128	3,120	3,100	3,102	3,111	3,105	3,104

Cont., Control; HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*.

<sup>a</sup>Finisher vitamin premix supplied per kg of diet: vitamin A IU: 7,000, vitamin D3 IU: 2,500, vitamin E: 20 mg, vitamin K: 3.0 mg, vitamin B1: 1.8 mg, vitamin B2: 5.0 mg, vitamin B6: 3.5 mg, vitamin B12: 0.012 mg, niacin: 30 mg, pantothenic acid: 12 mg, folic acid: 1.0 mg, biotin: 0.20 mg.

<sup>b</sup>Finisher minerals premix: manganese: 100 mg, zinc: 80 mg, iron: 60 mg, copper: 8.0 mg, selenium: 0.240 mg, iodine: 1 mg.

<sup>c</sup>Metabolizable energy was calculated by following Ravindran et al. (2014) while all other ingredients had been analyzed. HI, *Hermetia illucens*; AD, *Acheta domesticus*.

TABLE 5 Impacts of *H. illucens* and *A. domesticus* dietary groups on the productive performances of broilers.

Item	Control	<i>Hermetia illucens</i>			<i>Acheta domesticus</i>			PSEM	<i>p</i> -value				
		HI4	HI8	HI12	AD4	AD8	AD12		ANOVA	HI Lin.	HI Quad.	AD Lin.	AD Quad.
Live weight, g													
DOC	38.88	39.19	38.98	39.56	39.00	40.03	39.38	0.78	0.714	0.326	0.876	0.648	0.743
10 d	246.16 <sup>g</sup>	258.60 <sup>f</sup>	276.85 <sup>d</sup>	303.33 <sup>b</sup>	262.67 <sup>e</sup>	297.80 <sup>c</sup>	318.83 <sup>a</sup>	5.53	<0.001	<0.001	<0.001	<0.001	<0.001
24 d	1,027.33 <sup>cd</sup>	969.33 <sup>d</sup>	1,079.83 <sup>bc</sup>	1,170.83 <sup>ab</sup>	1,071.16 <sup>bc</sup>	1,105.67 <sup>bc</sup>	1,218.5 <sup>a</sup>	20.69	<0.001	0.014	0.574	<0.001	<0.001
35 d	1,901.03 <sup>f</sup>	1,960.95 <sup>e</sup>	2,059.75 <sup>c</sup>	2,158.90 <sup>a</sup>	1,974.15 <sup>d</sup>	2,126.44 <sup>b</sup>	2,151.77 <sup>a</sup>	21.39	<0.001	<0.001	<0.001	<0.001	<0.001
Average daily weight gain, g													
1–10 d	24.62 <sup>g</sup>	25.86 <sup>f</sup>	27.68 <sup>d</sup>	30.33 <sup>b</sup>	26.26 <sup>e</sup>	29.78 <sup>c</sup>	31.88 <sup>a</sup>	0.55	<0.001	<0.001	<0.001	<0.001	<0.001
11–24 d	55.79 <sup>bc</sup>	49.46 <sup>c</sup>	54.84 <sup>bc</sup>	62.35 <sup>ab</sup>	60.21 <sup>ab</sup>	58.65 <sup>ab</sup>	64.26 <sup>a</sup>	1.27	0.007	0.039	0.685	<0.001	<0.001
25–35 d	79.42 <sup>b</sup>	80.10 <sup>b</sup>	84.84 <sup>b</sup>	98.88 <sup>a</sup>	78.95 <sup>b</sup>	86.87 <sup>b</sup>	99.12 <sup>a</sup>	2.04	<0.001	0.002	0.907	<0.001	<0.001
1–35 d	53.28 <sup>e</sup>	54.87 <sup>d</sup>	58.75 <sup>c</sup>	61.35 <sup>a</sup>	55.14 <sup>d</sup>	59.67 <sup>b</sup>	60.32 <sup>b</sup>	0.65	<0.001	<0.001	0.114	<0.001	<0.001
Daily feed intake, g													
1–10 d	23.45	22.28	23.62	22.30	22.86	23.37	22.3	0.38	0.312	0.297	0.875	0.297	0.612
11–24 d	95.83	92.61	95.84	95.88	94.66	94.66	92.26	1.23	0.089	0.265	0.089	0.135	0.218
25–35 d	176.28	174.67	174.47	177.67	178.33	178.67	178.00	2.43	0.167	0.344	0.125	0.304	0.592
1–35 d	98.52	96.53	97.98	98.62	98.62	98.90	97.52	1.17	0.227	0.507	0.126	0.590	0.295
Feed conversion ratio, g/g													
1–10 d	0.95 <sup>a</sup>	0.86 <sup>b</sup>	0.85 <sup>b</sup>	0.78 <sup>c</sup>	0.87 <sup>b</sup>	0.73 <sup>bc</sup>	0.69 <sup>c</sup>	0.02	<0.001	<0.001	0.325	<0.001	0.001
11–24 d	1.99 <sup>a</sup>	1.74 <sup>ab</sup>	1.57 <sup>bc</sup>	1.72 <sup>bc</sup>	1.57 <sup>bc</sup>	1.52 <sup>bc</sup>	1.43 <sup>c</sup>	0.05	0.008	0.032	0.652	<0.001	0.041
25–35 d	2.26 <sup>a</sup>	2.18 <sup>ab</sup>	2.09 <sup>ab</sup>	1.80 <sup>c</sup>	2.21 <sup>ab</sup>	2.05 <sup>b</sup>	1.78 <sup>c</sup>	0.04	<0.001	<0.001	0.741	<0.001	0.002
1–35 d	1.63 <sup>a</sup>	1.54 <sup>b</sup>	1.54 <sup>b</sup>	1.42 <sup>c</sup>	1.56 <sup>b</sup>	1.45 <sup>c</sup>	1.41 <sup>c</sup>	0.02	<0.001	<0.001	0.405	<0.001	<0.001

DOC, day-old chick; HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*; PSEM, pooled standard error of the mean; ANOVA, analysis of variance; orthogonal polynomial contrast. HI lin., *H. illucens* linear; HI quad., *H. illucens* quadratic; AD lin., *A. domesticus* linear; AD quad., *A. domesticus* quadratic. Values marked with different superscripts within a row differed significantly ( $p < 0.05$ ).



The *H. illucens* meals affected FCR in a way across the feeding periods. Likewise, there were linear and quadratic effects in FCR across the intervals among the *A. domesticus* meals.

## 3.2 Hematology

### 3.2.1 Complete blood count

Hematology and serum biochemistry traits are mentioned in Table 6. The hematological parameters showed statistical differences ( $p < 0.05$ ) among all the treatments except for monocytes ( $p > 0.05$ ). The maximum hemoglobin (Hb), red blood cells (RBCs), hematocrits (HCT), platelets, total leucocytes (TLC), and lymphocytes (Lyn) were registered in 12% *A. domesticus* and 12% *H. illucens* and the minimum values were registered in 4% *H. illucens*. Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) were the highest in 12% *A. domesticus* and the lowest in the control group. HB, HCT, MCV, MCH, MCHC, TLC, heterocysts, and lymphocytes showed linear and quadratic responses, while a quadratic response to the *H. illucens* meals was found in RBCs and platelets. Similarly, HB, HCT, MCV, MCH, MCHC, platelets, and TLC were affected linearly and quadratically, while heterocysts and lymphocytes responded linearly to the *A. domesticus* meals.

### 3.2.2 Serum biochemistry

The serum biochemistry traits differed statistically ( $p < 0.05$ ) among the meals. Total protein, globulin, and albumin were the highest in the broilers fed 12% *A. domesticus*, while they were the lowest in the control meals. The maximum concentrations of uric acid, cholesterol, glucose, and creatinine were found in the control group, and the minimum concentrations were found in 12% *A. domesticus* meals. The orthogonal contrast depicted linear and quadratic responses in total protein, uric acid, cholesterol, glucose, and creatinine, while a quadratic effect in globulin and a linear effect in albumin were seen among *H. illucens* meals. Likewise, total protein, globulin, and creatinine registered a quadratic effect; albumin showed a linear response, while uric acid, cholesterol, and glucose presented linear and quadratic effects among *A. domesticus* meals.

## 3.3 Intestinal morphology

The dietary treatments, sites of the small intestine, and their interactions differed statistically ( $p < 0.05$ ) (Table 7). The Vh was the longest and similar at the jejunum and ileum when broilers were fed 12% *A. domesticus*, and at the jejunum site fed 12% *H. illucens*, 8% *H. illucens*, and 8% AD, while it was the shortest at the ileum site in the broilers fed the control diet and 4% *H. illucens* (Figure 1A). The highest Cd was observed at the jejunum in those fed the control diet and the lowest at the ileum site in those fed 12% *A. domesticus* (Figure 1B). The Vw was the largest and comparable at both sites when fed 12% *A. domesticus*, and at the jejunum site when fed 12% *H. illucens*, 8% *H. illucens*, and 8% *A. domesticus*, while it was the

smallest at the jejunum and ileum sites in the broilers fed the control diet, 4% *H. illucens*, and 4% *A. domesticus* and at the ileum site in the broilers fed 8% *H. illucens* and 8% *A. domesticus* (Figure 1C). Vh/Cd was the largest and similar at the jejunum and ileum sites in the broilers fed 12% *A. domesticus*, and at the jejunum site in the broilers fed 12% *H. illucens*, 8% *H. illucens*, and 8% *A. domesticus*. The lowest Vh/Cd ratios were recorded in the control and 4% *H. illucens* (Figure 1D).

## 3.4 Meat quality

Meat quality traits are presented in Table 8. These parameters differed significantly ( $p < 0.05$ ) among all the dietary treatments except for the drip loss, pH, shear force, and chroma ( $p > 0.05$ ). In the control treatment, maximum cooking loss and L\* were recorded, with the minimum found in the 12% *A. domesticus* meal. The a\* was the highest in 12% *A. domesticus* and the lowest in the control treatment. The b\* and H\* were the highest in 12% *A. domesticus* and 12% *H. illucens*, and the lowest in control. Cooking loss, a\*, and b\* showed linear and quadratic responses, while quadratic responses were found in L\* and H\* in the *H. illucens* groups. Likewise, *A. domesticus* meals exhibited a quadratic effect on cooking loss, and a linear effect on L\*, a\*, b\*, and H\* of the broilers.

## 4 Discussion

The findings showed that live weight and average daily weight gain were highest in the birds fed with diets containing 12% *A. domesticus* and 12% *H. illucens*. Constituents of the diets significantly impact the efficiency of the birds in poultry farming, particularly the protein content and amino acid profiles, which are crucial for growth performances. SBM and FM have lower protein contents and lower-quality amino acids compared to insects (Fisher et al., 2020; Hasnan et al., 2023). Insects such as *H. illucens* have well-balanced essential amino acids, with methionine 2.2% and lysine 6.1% (García-Vaquero and García, 2024; Hobbi et al., 2022), along with vitamins, while the SBM contains methionine 0.65% and lysine 2.95% (Akhtar and Isman, 2018). Moreover, insects offer superior fatty acids than SBM (Islam et al., 2022; Lu et al., 2022). Insects possess a well-balanced nutritional profile, including essential vitamins and minerals that promote robust and fast growth (Adli, 2021; Choi et al., 2023; da-Silva et al., 2024; Rehman et al., 2019). This study is aligned with those of Loponte et al. (2017), Pieterse et al. (2019), and Dabbou et al. (2018), indicating that substituting SBM at the levels of 10%, 15%, and 50% with BSF resulted in enhanced LW and ADG in Ross 308, Cobb 500, and barbery partridges. Many other previous studies by Biasato et al. (2018) and Vasilopoulos et al. (2023) have presented a substantial increase in live weight and daily weight gain in fast-growing Ross 708 and Ross 308 as well as intermediate-growing Hubbard hybrid when fed 10% to 15% *T. molitor* meals. Conversely, Ramos-Elorduy et al. (2002) and Biasato et al. (2016) did not report any impact on performances on fast-growing Arbor acres and Hubbard hybrid when fed *T. molitor* meals.

TABLE 6 Effects of *H. illucens* and *A. domesticus* meals on the hematology and serum biochemistry traits of broilers.

Trait	Control	<i>Hermetia illucens</i>			<i>Acheta domesticus</i>			PSEM	<i>p</i> -value				
		HI4	HI8	HI12	AD4	AD8	AD12		ANOVA	HI Lin.	HI Quad.	AD Lin.	AD Quad.
<b>Hematology</b>													
HB	10.00 <sup>c</sup>	8.33 <sup>f</sup>	8.90 <sup>e</sup>	11.20 <sup>b</sup>	9.76 <sup>c</sup>	9.90 <sup>c</sup>	12.30 <sup>a</sup>	0.88	<0.001	<0.001	0.027	<0.001	<0.001
RBCs	3.13 <sup>b</sup>	2.57 <sup>c</sup>	2.87 <sup>bc</sup>	3.67 <sup>a</sup>	2.83 <sup>bc</sup>	3.03 <sup>b</sup>	3.56 <sup>a</sup>	0.28	<0.001	0.175	0.041	0.020	0.003
HCT	30.07 <sup>e</sup>	26.63 <sup>f</sup>	35.73 <sup>b</sup>	39.56 <sup>a</sup>	35.30 <sup>c</sup>	32.36 <sup>d</sup>	39.53 <sup>a</sup>	1.19	<0.001	<0.001	<0.001	<0.001	<0.001
MCV	83.17 <sup>f</sup>	89.79 <sup>e</sup>	109.27 <sup>c</sup>	113.83 <sup>b</sup>	98.06 <sup>d</sup>	109.36 <sup>c</sup>	115.60 <sup>a</sup>	2.60	<0.001	<0.001	<0.001	0.005	<0.001
MCH	25.73 <sup>f</sup>	29.50 <sup>e</sup>	34.23 <sup>c</sup>	36.13 <sup>b</sup>	31.76 <sup>d</sup>	34.23 <sup>c</sup>	37.66 <sup>a</sup>	0.98	<0.001	0.002	<0.001	<0.001	<0.001
MCHC	30.30 <sup>d</sup>	32.73 <sup>e</sup>	33.33 <sup>b</sup>	33.67 <sup>b</sup>	32.43 <sup>e</sup>	33.23 <sup>b</sup>	35.33 <sup>a</sup>	0.64	<0.001	0.017	<0.001	<0.001	<0.001
Platelets	24,000.00 <sup>c</sup>	13,033.33 <sup>f</sup>	13,966.67 <sup>e</sup>	24,966.67 <sup>b</sup>	14,666.66 <sup>e</sup>	16,666.67 <sup>d</sup>	27,000.00 <sup>a</sup>	1,226	< 0.001	0.083	<0.001	<0.001	<0.001
TLC	18,066.67 <sup>c</sup>	7,542.28 <sup>f</sup>	11,000.00 <sup>d</sup>	21,000.00 <sup>b</sup>	10,066.37 <sup>e</sup>	17,466.67 <sup>c</sup>	23,000.00 <sup>a</sup>	1,275	< 0.001	<0.001	<0.001	<0.001	<0.001
Heter.	36.67 <sup>d</sup>	40.19 <sup>c</sup>	53.33 <sup>ab</sup>	52.66 <sup>ab</sup>	44.33 <sup>c</sup>	50.33 <sup>b</sup>	55.67 <sup>a</sup>	1.69	<0.001	<0.001	0.046	0.027	0.549
Lym.	44.67 <sup>b</sup>	33.67 <sup>d</sup>	40.59 <sup>c</sup>	56.33 <sup>a</sup>	40.66 <sup>c</sup>	43.33 <sup>bc</sup>	56.66 <sup>a</sup>	1.80	<0.001	<0.001	0.003	<0.001	0.084
Mono.	1.99	1.99	2.00	2.00	1.99	1.99	2.00	0.14	0.469	0.678	0.841	0.932	0.459
<b>Serum biochemistry</b>													
T. protein	2.73 <sup>c</sup>	2.83 <sup>bc</sup>	3.20 <sup>ab</sup>	3.33 <sup>a</sup>	3.23 <sup>ab</sup>	3.13 <sup>abc</sup>	3.36 <sup>a</sup>	0.06	0.001	0.004	0.002	0.511	0.03
Glob.	1.13 <sup>c</sup>	1.33 <sup>b</sup>	1.90 <sup>ab</sup>	1.93 <sup>ab</sup>	1.43 <sup>b</sup>	1.63 <sup>ab</sup>	2.16 <sup>a</sup>	0.03	0.006	0.639	0.011	0.077	0.009
Alb.	1.20 <sup>c</sup>	1.30 <sup>bc</sup>	1.50 <sup>ab</sup>	1.50 <sup>ab</sup>	1.40 <sup>abc</sup>	1.30 <sup>bc</sup>	1.60 <sup>a</sup>	0.07	0.003	0.015	0.789	0.002	0.487
Uric acid	4.13 <sup>a</sup>	4.13 <sup>a</sup>	3.93 <sup>ab</sup>	3.53 <sup>bc</sup>	3.63 <sup>bc</sup>	3.86 <sup>abc</sup>	3.43 <sup>c</sup>	0.06	0.004	0.035	0.042	0.011	0.042
Cholesterol	154.33 <sup>a</sup>	152.67 <sup>a</sup>	140.33 <sup>b</sup>	124.33 <sup>c</sup>	152.67 <sup>a</sup>	136.33 <sup>b</sup>	120.33 <sup>c</sup>	2.92	<0.001	<0.001	<0.001	<0.001	<0.001
Glucose	212.67 <sup>a</sup>	212.33 <sup>a</sup>	201.33 <sup>b</sup>	162.67 <sup>d</sup>	175.33 <sup>c</sup>	196.33 <sup>b</sup>	135.67 <sup>e</sup>	2.78	<0.001	<0.001	<0.001	<0.001	<0.001
Creatinine	0.53 <sup>a</sup>	0.53 <sup>a</sup>	0.43 <sup>ab</sup>	0.33 <sup>bc</sup>	0.40 <sup>abc</sup>	0.43 <sup>ab</sup>	0.23 <sup>c</sup>	0.03	0.007	0.011	0.011	0.697	0.008

Traits are as follows: Hb g/dL, hemoglobin; RBCs  $\times 10^6/\mu\text{L}$ , red blood cells; HCT %, hematocrits; MCV fL, mean corpuscular volume; MCH pg, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; Platelets/ $\mu\text{L}$ ; TLC  $\times 10^3/\mu\text{L}$ , total leucocytes; Heter. (%), heterophils; Lym. (%), lymphocytes; Mono. (%), monocytes; T. protein g/dL, total protein; Glob. g/dL, globulin; Alb. g/dL, albumin; Uric acid g/dL; Cholesterol mg/dL; Glucose mg/dL; Creatinine mg/dL. HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*; PSEM, pooled standard error of the mean; ANOVA, analysis of variance; orthogonal polynomial contrast; HI lin., *H. illucens* linear; HI quad., *H. illucens* quadratic; AD lin., *A. domesticus* linear; SF quad., *A. domesticus* quadratic. Values marked with different superscript within a row were significantly different ( $p < 0.05$ ).

TABLE 7 The impact of dietary treatments, intestinal sites, and their interactions on the gut morphometric indices.

Index	Fixed factors	df	F-value	p-value
Villus height (µm)	Meals	6	58.91	<0.001
	Intestinal sites	1	187.21	<0.001
	Meals × intestinal sites	6	4.11	0.002
Villus width (µm)	Meals	6	5.32	<0.001
	Intestinal sites	1	37.33	<0.001
	Meals × intestinal sites	6	17.16	<0.001
Crypt depth (µm)	Meals	6	156.54	<0.001
	Intestinal sites	1	406.19	<0.001
	Meals × intestinal sites	6	20.74	<0.001
Villus height/crypt depth (µm)/(µm)	Meals	6	3.42	0.006
	Intestinal sites	1	54.67	<0.001
	Meals × intestinal sites	6	3.50	0.002

In the present work, daily feed intake did not change significantly among diets. However, feed intake typically increases when the protein and lipid contents decrease in the feed (Van Harn et al., 2019). The feed color might also affect the feed conversion ratio (Hartinger et al., 2021). Chu et al. (2020), Dabbou et al. (2021), and Sajjad et al. (2024c) registered comparable results in broiler Ross 308.

In this study, the feed conversion ratio was improved when broilers were fed 12% *A. domesticus* and 12% *H. illucens*. The feed

conversion ratio is the most imperative indicator of meal efficiency in livestock production (Davison et al., 2023; Prakash et al., 2020). It can decrease feed conversion ratio by replacing SBM with insect meal owing to the highest nutrients, balanced AAs, palatability, digestibility, and lower anti-nutritional aspects (Parrini et al., 2023; Siddiqui et al., 2024). This study’s results are analogous to the findings of Schiavone et al. (2017), who indicated that replacing soybean oil with *H. illucens*, either partially or completely, enhanced the feed conversion ratio in Ross 308. The study by Kim et al. (2020) and Lalev

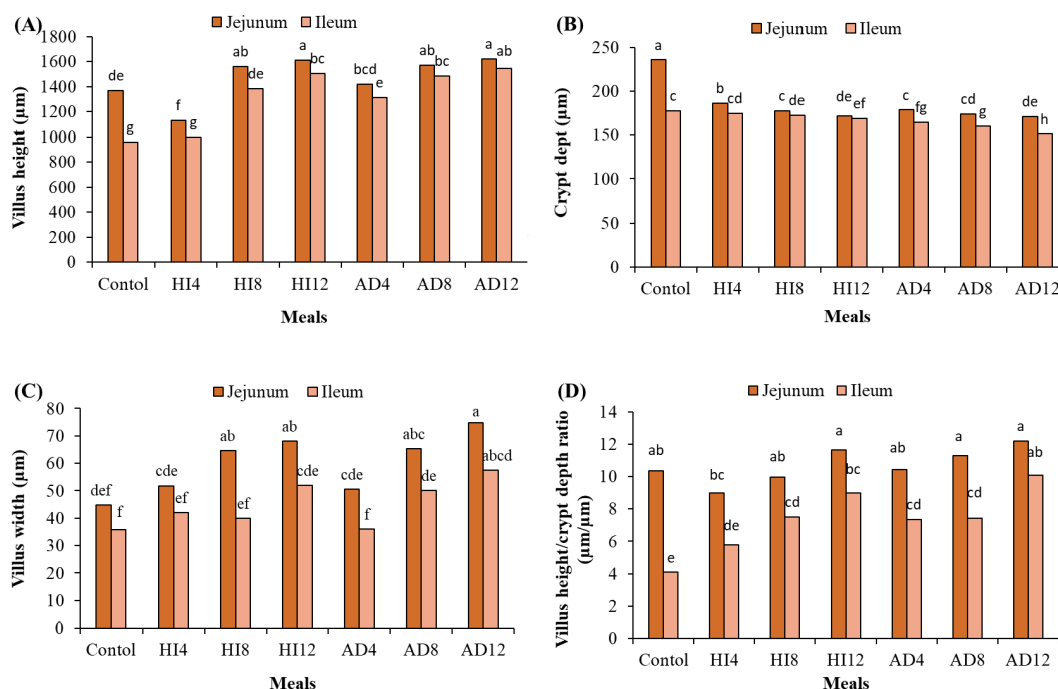


FIGURE 1 Interactions between intestinal sites and dietary treatments; villus height (A), crypt depth (B), villus width (C), and villus height/crypt depth ratio (D). Values marked with different letters indicate statistically significant differences (p < 0.05).

TABLE 8 Impact of *H. illucens* and *A. domesticus* meals on the meat quality of broilers.

Trait	Control	<i>Hermetia illucens</i>			<i>Acheta domesticus</i>			PSEM	p-value				
		HI4	HI8	HI12	AD4	AD8	AD12		ANOVA	HI lin.	HI Quad.	AD Lin.	AD Quad.
Meat pH	6.08	6.11	6.10	6.10	6.10	6.12	6.08	0.57	0.490	0.539	0.753	0.837	0.528
Cooking loss	33.32 <sup>a</sup>	29.32 <sup>b</sup>	26.12 <sup>c</sup>	22.79 <sup>d</sup>	33.16 <sup>a</sup>	30.08 <sup>b</sup>	18.70 <sup>c</sup>	1.13	<0.001	<0.001	0.002	0.278	<0.001
Drip loss	2.35	2.39	2.32	2.29	2.27	2.34	2.39	0.24	0.967	0.482	0.693	0.537	0.472
Share-force	61.02	61.02	60.84	61.06	61.04	61.07	60.70	1.48	0.359	0.643	0.233	0.983	0.467
Lightness L*	58.69 <sup>a</sup>	54.70 <sup>ab</sup>	49.62 <sup>b</sup>	48.94 <sup>b</sup>	57.18 <sup>a</sup>	55.82 <sup>ab</sup>	50.84 <sup>bc</sup>	1.25	0.026	0.447	<0.001	0.002	0.337
Redness a*	12.36 <sup>c</sup>	16.29 <sup>ab</sup>	16.02 <sup>ab</sup>	14.49 <sup>b</sup>	15.95 <sup>ab</sup>	16.13 <sup>ab</sup>	16.99 <sup>a</sup>	0.89	0.015	0.042	<0.001	0.009	0.385
Yellowness b*	12.96 <sup>c</sup>	15.32 <sup>bc</sup>	14.73 <sup>cd</sup>	23.40 <sup>a</sup>	17.43 <sup>bc</sup>	16.03 <sup>ab</sup>	18.45 <sup>a</sup>	0.76	0.002	<0.001	<0.001	<0.001	0.517
Chroma C*	24.93 <sup>a</sup>	24.62 <sup>a</sup>	24.70 <sup>a</sup>	24.75 <sup>a</sup>	24.70 <sup>a</sup>	24.96 <sup>a</sup>	24.93 <sup>a</sup>	1.04	0.195	0.362	0.104	0.346	0.992
Hue H*	23.29 <sup>b</sup>	47.50 <sup>ab</sup>	43.72 <sup>ab</sup>	56.17 <sup>a</sup>	40.89 <sup>ab</sup>	44.77 <sup>ab</sup>	58.21 <sup>a</sup>	1.44	0.134	0.633	<0.001	<0.001	0.990

HI4, 4% *H. illucens*; HI8, 8% *H. illucens*; HI12, 12% *H. illucens*; AD4, 4% *A. domesticus*; AD8, 8% *A. domesticus*; AD12, 12% *A. domesticus*. Values marked with different superscript within a row were significantly different ( $p < 0.05$ ). PSEM, pooled standard error of the mean; ANOVA, analysis of variance; orthogonal polynomial contrast; HI lin., *H. illucens* linear; HI quad., *H. illucens* quadratic; AD lin., *A. domesticus* linear; SF quad., *A. domesticus* quadratic.

et al. (2022) revealed that the inclusion of 5% and 10% *H. illucens* and *B. mori* in broiler meals led to significant improvement in feed conversion ratio in broilers. Bovera et al. (2015) and Kierończyk et al. (2018) found an improvement in the feed conversion ratio in Ross 308 feeding 10% to 30% *T. molitor*. Ognik et al. (2020) reported that 15% *H. illucens* improved FCR in young turkeys, while Murawska et al. (2021) found no improvement in feed conversion ratio in Ross 308, even at 75% *H. illucens*.

In this investigation, 12% *A. domesticus* and 12% *H. illucens* resulted in the highest Hb, RBCs, HCT, MCV, MCH, MCHC, platelets, total leucocytes, and lymphocytes. Insects have a superior nutritional profile compared to traditional protein sources, which may impact the hematology of the broilers (Marono et al., 2017; Zulkifli et al., 2022). They contain higher concentrations of essential amino acids, absorbable minerals (iron and zinc), and functional lipids, which can boost blood traits by promoting RBC production, hemoglobin levels, and the overall health of the birds (Slimen et al., 2023; Zhou et al., 2022). The higher iron content in insect meal is vital for the synthesis of hemoglobin and erythropoiesis in the broiler (Elliott, 2008; Sajjad et al., 2024a). Zinc is another mineral abundant in insects that plays an essential role in enzymatic functions and the development of immune cells, potentially enhancing leukocyte production and strengthening the immune system (Vasilopoulos et al., 2024a). This study's results are consonant with Biasato et al. (2017), who noticed that hematological traits improved in broilers Ross 708 when fed insect-based meals. Gariglio et al. (2019), Kim et al. (2020), Schiavone et al. (2017), and Ognik et al. (2020) depicted that supplanting 4% to 100% soybean oil or SBM with insects did not affect the hematological attributes. Lymphocytes were increased when the broiler was fed 20% *H. illucens* (De Souza Vilela et al., 2021).

In this inquiry, the diets containing 12% *A. domesticus* and 12% *H. illucens* exhibited the lowest concentrations of creatinine, uric acid, glucose, and cholesterol, while showing the highest levels of total protein, albumin, and globulin. The chitin and chitosan of

insect exoskeletons could potentially have chelating effects that lower the concentrations of uric acid, glucose, and cholesterol in the blood (Hossain and Blair, 2007; Khambualai et al., 2008). This, in turn, may enhance the metabolic functioning of the bird (Alagawany et al., 2022; Sypniewski et al., 2020). The nutritional value of the feed may strengthen the immunity system of birds and increase their resistance against several infectious diseases (Schiavone et al., 2017). Total protein and globulin levels are usually influenced by isoenergetic and isoproteic diets (Bovera et al., 2016). This study's results are consistent with previous findings by Sedgh-Gooya et al. (2021) and Bovera et al. (2018), who revealed that 5% to 100% inclusion of *T. molitor* meals increased the concentrations of total protein and globulin and diminished the level of uric acid in Shaver brown broilers and Bovans white laying hens.

In the current study, the interactions between the dietary treatment and the sites of the small intestine showed significant differences. The villus height, villus width, and villus height/crypt depth ratio were the largest at the jejunum and ileum sites of the intestine using 12% *A. domesticus* and 12% *H. illucens* with the smallest crypt depth. The morphology of the intestine is affected by the consumption of insects, which have a unique composition of bio-active compounds, including chitin, antimicrobial peptides, and short-chain fatty acids (Vasilopoulos et al., 2024b). Moreover, chitin may act as a prebiotic that stimulates beneficial gut microbiota such as *Lactobacillus* and *Bifidobacterium*, enhancing the gastrointestinal tract's health and villus growth (Mohan et al., 2023). Furthermore, antimicrobial peptides might contribute to maintaining intestinal integrity by decreasing the load of pathogenic bacteria, which decreases inflammation and promotes healthy villus development (Patyra and Kwiatek, 2023). Nutrient absorption is primarily related to histological indices of the intestine, particularly the villi and crypts (Amer et al., 2021). The longest villi and the shortest crypts play a fundamental role in the efficient digestion and absorption of

nutrients (Ravindran and Abdollahi, 2021). This study's results are aligned with Sajjad et al. (2024a) and Sajjad et al. (2024c), who found comparable effects in Ross 308 when fed 12% *Spodoptera frugiperda* and 12% *A. diaperinus*. Another study by Dabbou et al. (2018) and Dabbou et al. (2021) reported that the gut histological indices improved in broilers feeding on defatted and modified *H. illucens* meals.

The findings of this study presented that 12% *A. domesticus* and 12% *H. illucens* diets had the maximum a\*, b\*, and H\* values, along with the minimum cooking loss and L\* values. The physical meat and skin properties, such as color and tenderness, do not differ, as color can affect consumer acceptance (Bovera et al., 2016; Yang et al., 2011). High concentrations of lauric acid, myristic acid, and eicosapentaenoic acid, as well as the pigments of the feed ingredients, can influence the color and texture of meat (Cullere et al., 2019; Vilela et al., 2021). Replacing SBM (7.8%) with *H. illucens* reduced cooking loss in broiler Ross 308 (Leiber et al., 2017). Murawska et al. (2021), Kim et al. (2019), and Sajjad et al. (2024a) found that increasing the levels of *H. illucens* and lesser mealworm *A. diaperinus* in the feeds dwindled cooking loss while increasing the b\* (Ross 308). However, several factors, including feed, stage, breed, and slaughtering techniques, can significantly affect the meat (Mir et al., 2017).

Incorporating local insect species as feed ingredients in the poultry sector offers several advantages. Utilizing locally sourced insects can reduce dependency on imported feed ingredients, lower feed costs, and contribute to a more sustainable and environmentally friendly poultry industry. Additionally, insect farming has a smaller carbon footprint, minimizes waste, and promotes circular economies (Madau et al., 2020).

## 5 Conclusions

Utilizing *H. illucens* and *A. domesticus* as a protein source significantly impacted live weight, average body weight gain, feed conversion ratio, hematological traits, gut histology, and meat quality in Ross 308. Overall, the birds fed with 12% *H. illucens* and 12% *A. domesticus* for 35 days performed better than those fed other diets. Therefore, it can be concluded that *H. illucens* and *A. domesticus* are viable alternative ingredients that can substitute SBM to develop a local-based feed ingredient for broiler chickens at the optimal inclusion rate of 12% without compromising performance and health as evidenced by blood parameters, gut histology, and meat quality of the birds. Future research should investigate how *H. illucens* and *A. domesticus* meals affect the gut microbiota of broilers when utilized as a principal protein source in poultry feed.

## Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding authors.

## Ethics statement

The animal study was approved by University of Animal and Veterinary Sciences (UVAS), Ravi Campus, Pattoki, Lahore, Pakistan. The Ethical Review Committee (No. DR/495) at UVAS approved all the procedures. The study was conducted in accordance with the local legislation and institutional requirements.

## Author contributions

FM: Conceptualization, Formal analysis, Investigation, Methodology, Writing – original draft. AS: Conceptualization, Data curation, Formal analysis, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. MS: Data curation, Formal analysis, Methodology, Software, Validation, Visualization, Writing – original draft. MA: Formal analysis, Investigation, Software, Validation, Writing – original draft. HB: Conceptualization, Data curation, Validation, Visualization, Writing – original draft. MGA: Data curation, Methodology, Writing – original draft. MB: Conceptualization, Resources, Writing – review & editing. RM: Funding acquisition, Writing – review & editing.

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## Conflict of interest

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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