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Research article

# Feeding trial on the effects of high-protein, low-energy diets on lean meat production in broiler

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## Abstract

This study aimed to produce lean meat from broiler chickens fed a high-protein, low-energy diet. For 35 days, 250 straight-run Cobb-500 chicks were arbitrarily assigned to five dietary groups, with 50 birds in each group and 10 birds as replicates per cage. The per-kg diets of the different groups contained 1. Twenty-two percent crude protein (CP), 2,900 kcal metabolizable energy (ME), 2. Twenty-two percent CP and 2,600 kcal ME, 3. Twenty-four percent CP and 2,600 kcal ME, 4**.** Twenty-six percent CP and 2,600 kcal ME, and 5. Twenty-eight percent CP and 2,600 kcal ME. On the final day, 15 birds from each group were sacrificed to evaluate carcass traits, meat colour, gastrointestinal tract pH, and blood profiles. Group 3 (24% CP and 2,600 kcal ME) achieved a substantially higher weight (1,500 g), consumed more feed (2,445 g), and had a better FCR (1.67) than the other groups ($P < 0.05$). pH levels tended to be higher in the duodenum in Group 5, whereas Group 3 showed optimal serum albumin, urea, and calcium levels. Compared to the other groups, Group 3 had a higher CP content and redder meat. The dressing yields (without skin) were 58.47, 58.51, 62.06, 59.49, and 60.03% in Groups 1, 2, 3, 4, and 5, respectively, with Group 3 having the highest value ($P < 0.05$). Muscle fat was lower in Groups 2 and 3, but skin fat was lower in Groups 3 and 4 ($P < 0.05$), indicating that the possibility of obtaining lean meat was higher in Group 3. Therefore, diets containing 24% CP and 2,600 kcal ME/kg are recommended to farmers for better growth and meat quality (leanness, redness, and low abdominal fat) of broilers.

**Keywords**: Broilers, High-Protein, Low-Energy Diet, Lean Meat Production.

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# INTRODUCTION

The issue of higher abdominal fat and lower dressed yield in broilers has been a major concern for both producers and consumers (Połtowicz et al., 2015), because abdominal fat is related to the health of consumers, and production costs are related to the profitability of farmers. Consumers prefer low-fat, safe, and good-quality meat at affordable prices, along with a higher dressed yield from broilers. Scientists have investigated feed additives such as organic acids, enzymes, probiotics, phytobiotics, and different combinations of various ingredients to reduce abdominal fat and improve dressed yield, lean meat, and profitability in broilers (Hoque et al., 2022, Rahman et al., 2022; Chowdhury et al., 2024). Dietary manipulation, such as a high-protein, low-energy diet, may be a possible solution for reducing costs and improving the dress yield and meat quality of poultry (Peng et al., 2023).

Lowering both protein and energy levels reduces the protein content in muscle but increases it due to the higher energy content in the diet (Marcu et al., 2013). Similarly, lowering the protein content in the diet to as low as 3.0% reduces growth and the carcass composition becomes inferior in broilers, even when all known nutrient requirements are met (Aletor et al., 2000; Bregendahl et al., 2002; Sterling et al., 2005; Waldroup et al., 2005). It is also well-documented that dietary composition and the ratio of macronutrients have a major effect on the performance and body composition of broilers (Nieto et al., 1997; Collin et al., 2003).

Energy is essential for the homeostasis, growth, and movement of birds. Overconsumed energy is stored as glycogen and abdominal fat, but increases feed costs and lowers meat quality (Mir et al., 2017). Another study found that a high-energy diet with standard protein levels do not affect broiler feed intake or weight gain (Salinas-Chavira et al., 2016). In addition, broilers fed low-energy diets with low-amino acids displays similar growth performance to those fed high-energy diets (Salinas-Chavira et al., 2016). Therefore, protein and energy levels are important for obtaining lean meat, and farmers' profitability should be considered by many researchers.

Breeding companies recommend a diet for broilers that promotes higher weight gain with relatively low feed intake, which is often associated with increased fat content in the muscle, skin, and abdominal areas (Redoy et al., 2021). However, reducing fat in muscle and skin is desirable for producing leaner meat from broilers, whereas feed conversion efficiency remains crucial for farmer profitability. This study differs from the earlier ones by optimizing both the levels of protein and energy in the broiler diet for efficient lean meat production with reduced fat in muscles, skin, and abdomen. Some novel insights have also been provided regarding how these dietary manipulations affect meat redness and fat content, along with serum biochemical indices, which are useful and applicable recommendations for broiler raising in cost-sensitive agricultural settings.

# MATERIALS AND METHODS

## Experimental birds

The feeding trial and sample analyses were performed in accordance with the rules and regulations of the Animal Welfare and Experimentation Ethics Committee (AWEEC/BAU/2023/18). A total of 250 straight-run Cobb-500 day-old broilers chicks with an average body weight of 44.0 ± 1.15 g were randomly assigned into five dietary groups, where each group consisted of 50 birds as replicates in 5 cages (10 birds per cage) considering a completely randomized design (CRD). The dietary groups comprised 1. Twenty-two percent CP and 2,900 kcal ME/kg feed; 2. Twenty-two percent CP and 2,600 kcal ME /kg feed; 3. Twenty-four percent CP and 2,600 kcal ME/kg feed; 4. Twenty-six percent CP and 2,600 kcal ME/kg feed; and 5. Twenty-eight percent CP and 2,600 kcal ME/kg feed. A single diet was offered to each group throughout the experimental period to avoid the effects of feeding in different phases because the experiment was conducted in a completely randomized design. The formulations and chemical constituents of the diets are listed in Table 1.

**Table 1** Feed ingredients and chemicals composition of diets

| Ingredients (%) | Dietary groups (% CP + ME kcal/kg feed) | | | | |
|---|---|---|---|---|---|
| | 1 (22+2900) | 2 (22+2600) | 3 (24+2600) | 4 (26+2600) | 5 (28+2600) |
| Corn | 52.20 | 48.30 | 46.50 | 44.20 | 37.30 |
| Wheat bran | 0.50 | 2.00 | 2.60 | 1.00 | 2.00 |
| De-oiled rice bran | 1.30 | 9.50 | 4.30 | 1.50 | 1.00 |
| Soybean meal | 38.40 | 35.30 | 42.00 | 48.60 | 54.10 |
| Soybean oil | 3.30 | 0.40 | 0.10 | 0.20 | 1.30 |
| Limestone | 1.00 | 1.20 | 1.20 | 1.20 | 1.00 |
| Oyster shell | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| Di-calcium phosphate | 1.50 | 1.50 | 1.50 | 1.50 | 1.50 |
| Mineral mixture | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| *Vitamin-mineral premix | 0.45 | 0.45 | 0.45 | 0.45 | 0.45 |
| Choline chloride | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 |
| Methionine | 0.20 | 0.20 | 0.20 | 0.20 | 0.20 |
| Salt | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| **Chemical composition (% on fed basis)** | | | | | |
| Crude protein | 22.03 | 22.01 | 24.03 | 26.05 | 28.01 |
| Calcium | 0.94 | 1.02 | 1.02 | 1.02 | 0.97 |
| Available phosphorous | 0.38 | 0.40 | 0.40 | 0.41 | 0.43 |
| Lysine | 1.28 | 1.26 | 1.42 | 1.58 | 1.73 |
| Methionine | 0.57 | 0.56 | 0.59 | 0.62 | 0.65 |
| ME (kcal/kg feed) | 2900 | 2610 | 2602 | 2607 | 2607 |

1 = 22% CP + 2,900 kcal ME/kg feed; 2 = 22% CP + 2,600 kcal ME/kg feed; 3 = 24% CP + 2,600 kcal ME/kg feed; 4 = 26% CP + 2,600 kcal ME/kg feed; 5 = 28% CP+ 2,600 kcal ME/kg feed; *Nutrient content of premix per kg diet: vitamin A palmitate = 6,600 IU; cholecalciferol = 2,200 IU; menadione dimethylpyridine bisulfite = 2.2 mg; riboflavin = 4.4 mg; pantothenic acid = 13 mg; niacin = 40 mg; choline chloride = 500 mg; biotin = 1 mg; vitamin B12 = 22 µg; ethoxyquin = 125 mg; iron = 50 mg; copper = 6 mg; zinc = 40 mg; manganese = 60 mg; selenium = 0.2 mg.

## Management of birds

The birds were reared in an intensive floor pen with 1.5–2.0 inches of sawdust as bedding, ensuring 11 square feet per replication (1.10 square feet per bird). The litter was agitated weekly to avoid the formation of ammonia and every two weeks the top layer of litter containing droppings was replaced with fresh litter. The birds were provided feed and water *ad libitum* twice a day. The feeder was cleaned weekly and the drinker was cleaned twice daily. In the first week, the brooding temperature was maintained at 34°C and then decreased by 3°C each week until it reached 21°C. The standard lighting program followed the Cobb-500 commercial broiler management guidelines (Redoy et al., 2021). The birds were vaccinated on days 5 and 12 against Newcastle disease and infectious bursal disease, respectively.

## Data collection

Live weight, feed offered, and feed refusal were recorded weekly to calculate weekly weight gain, feed intake, and feed conversion ratio (FCR). Birds were observed twice daily for physical fitness, and if any symptoms of disease or dead birds were observed.

## Dressed yield, carcass traits, sampling, and analysis

At 35 days of age, 15 birds from each group were selected based on the average weight of the cage and euthanized via stunning. During slaughter, approximately 10 ml of blood samples from each bird were collected in a falcon tube. For serum separation, samples were centrifuged at 3,410 g for 15 minutes and stored at -20°C for further analysis (Rahman et al., 2022). Specific kits were used to measure serum total protein, albumin, urea, uric acid, creatinine, calcium, and phosphorus levels using a biochemistry analyzer (Urit-810, Urit Medical Electronic Group Co. Ltd., China). After complete bleeding, the feathers were plucked, and the viscera and giblets were removed from the carcass. Both live and slaughtered birds (after complete bleeding), along with their skin, breasts, thighs, and drumsticks were weighed. The dressing percentage of broilers was calculated as the proportion of the bird's live weight that resulted in usable meat after slaughter and the removal of feathers, head, feet, and internal organs; however, the skin was not included in this case. Additionally, fat deposits surrounding the proventriculus and gizzard extending to the cloaca were collected and classified as abdominal fat. Subcutaneous fat beneath the skin was collected, weighed, and designated as skin fat (Ferrini et al., 2008). Immediately after slaughter, the breasts were thoroughly wiped with soft tissue paper and lightness (L*), redness (a*), and yellowness (b*) were determined using a colorimeter (CR-410 colorimeter; Minolta, Japan) three times within 45 minutes of evisceration. The pH of the crop, duodenum, jejunum, ileum, caecum, and colon digesta were measured using a pH meter (PHS-25, ISO9000, Nanjing, China) at 25°C. The proximate components of the feed and meat samples were examined according to recommended methods (AOAC, 2005).

## Statistical analysis

A one-way analysis of variance was performed using SPSS version 22 (Inc., Armonk, NY, USA). Duncan's multiple range test was used to assess the significance of the means. The significance level for the mean comparison was set at a 5% significance level ($P < 0.05$). All data in the tables are presented as mean ± standard deviation.

# RESULTS

## Performance

Lowering the energy in the diet showed a tendency toward better weight gain in Group 2 than in Group 1 ($P > 0.05$; Table 2). Subsequently, significantly increased protein-containing diets ($P < 0.05$) resulted in greater body weight in Group 3 than in Group 1, but there were no significant positive changes in Groups 4 and 5. Diets containing less energy were consumed more than the feed for Group 1, which was significantly higher at 24%, 26%, and 28% protein levels in Groups 3, 4, and 5, respectively ($P < 0.05$). An important parameter is the feed intake/body weight gain (FCR), which was better in Group 3 (1.67), where birds were offered a 24% CP and 2,600 kcal ME/kg diet, than in other groups.

**Table 2** Effect of different level of energy and protein on growth performance of broilers

| Parameters | Dietary groups (% CP + ME kcal/kg feed) | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|
| | 1 (22+2900) | 2 (22+2600) | 3 (24+2600) | 4 (26+2600) | 5 (28+2600) | | |
| FBW (g/b) | $1225^{c}$±41 | $1363^{bc}$±108 | $1500^{a}$±30 | $1502^{a}$±58 | $1471^{b}$±103 | 32.84 | 0.004 |
| BWG (g/b) | $1187^{b}$±39 | $1325^{ab}$±73 | $1460^{a}$±31 | $1459^{a}$±61 | $1419^{ab}$±79 | 30.66 | 0.035 |
| FI (g/b) | $2022^{b}$±85 | $2288^{ab}$±41 | $2445^{a}$±42 | $2543^{a}$±41 | $2580^{a}$±95 | 56.26 | 0.002 |
| FCR | $1.70^{bc}$±0.04 | $1.73^{b}$±0.11 | $1.67^{c}$±0.06 | $1.74^{b}$±0.09 | $1.81^{a}$±0.03 | 0.020 | 0.003 |

1 = 22% CP + 2,900 kcal ME/kg feed; 2 = 22% CP + 2,600 kcal ME/kg feed; 3 = 24% CP + 2,600 kcal ME/kg feed; 4 = 26% CP + 2,600 kcal ME/kg feed; 5 = 28% CP+ 2,600 kcal ME/kg feed

$^{abc}$ Means values within the same row with different superscripts differed significantly ($P < 0.05$). FBW = Final body weight; BWG = Body weight gain; FI = Feed intake; FCR = Feed conversion ratio; g = Gram; b = Bird.

## pH of different segments of the digestive tract

The pH of the different segments of the digestive tract, except for the duodenum, was not affected by feeding birds a high-protein or low-fat diet (P > 0.05; Table 3). However, the pH of the lower duodenum was significantly higher in Groups 2 (5.90) and 3 (5.71) than in Groups 1 (6.04), 4 (6.08), and 5 (6.22) (P < 0.05).

**Table 3** Effect of different level of energy and protein on pH in different segments of the digestive tract of broilers

| Parameters | Dietary groups (% CP + ME kcal/kg feed) | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|
| | 1 (22+2900) | 2 (22+2600) | 3 (24+2600) | 4 (26+2600) | 5 (28+2600) | | |
| Crop | 5.25±0.3 | 5.21±0.3 | 5.38±0.6 | 5.47±0.5 | 5.53±0.4 | 0.10 | 0.66 |
| Duodenum | 6.04$^{a}$±0.5 | 5.90$^{b}$±1.0 | 5.71$^{b}$±1.1 | 6.08$^{a}$±0.8 | 6.22$^{a}$±0.9 | 0.20 | 0.00 |
| Jejunum | 6.29±1.2 | 6.52±1.1 | 6.06±1.5 | 6.29±0.8 | 6.43±0.3 | 0.24 | 0.88 |
| Ileum | 6.42±0.3 | 6.58±0.5 | 6.23±0.2 | 6.30±0.2 | 6.42±0.3 | 0.08 | 0.32 |
| Caecum | 6.82±0.1 | 6.52±0.4 | 6.39±0.2 | 6.91±0.3 | 6.99±0.4 | 0.09 | 0.33 |
| Colon | 5.25±0.3 | 5.21±0.3 | 5.38±0.6 | 5.47±0.5 | 5.53±0.4 | 0.10 | 0.66 |

1 = 22% CP + 2,900 kcal ME/kg feed; 2 = 22% CP + 2,600 kcal ME/kg feed; 3 = 24% CP + 2,600 kcal ME/kg feed; 4 = 26% CP + 2,600 kcal ME/kg feed; 5 = 28% CP+ 2,600 kcal ME/kg feed.
$^{ab}$ Means values within the same row with different superscripts differed significantly (P < 0.05).

## Serum protein indices and minerals status

Birds that received the high-protein and low-energy diets showed inconsistent values for serum total protein, albumin, globulin, urea, and uric acid, whereas calcium levels increased with increasing protein levels (Table 4). In addition, phosphorus and creatinine concentrations were unaffected in birds fed low-fat and high-protein diets (P > 0.05). Birds in Group 5 showed the highest protein, albumin, and globulin levels compared to the other groups (P < 0.05), and this highest value was comparable with that of Group 1 (P > 0.05). However, higher calcium levels (mg/dl) were observed in Groups 4 (10.56), 3 (7.60), and 5 (12.32), while the highest urea and uric acid levels were observed in Group 5 than Group 1 (P < 0.05).

**Table 4** Effect of different level of energy and protein on serum protein indices and minerals status of broilers

| Parameters | Dietary groups (% CP + ME kcal/kg feed) | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|
| | 1 (22+2900) | 2 (22+2600) | 3 (24+2600) | 4 (26+2600) | 5 (28+2600) | | |
| Protein (g/dl) | 4.01$^{a}$±0.19 | 3.36$^{c}$±0.21 | 3.72$^{b}$±1.03 | 3.57$^{b}$±0.20 | 4.12$^{a}$±0.24 | 0.13 | 0.001 |
| Albumin (g/dl) | 1.10$^{c}$±0.33 | 1.23$^{b}$±0.19 | 2.55$^{a}$±0.03 | 1.19$^{c}$±0.17 | 1.32$^{b}$±0.11 | 0.15 | 0.003 |
| Globulin (g/dl) | 2.74$^{a}$±0.40 | 2.13$^{b}$±0.20 | 1.17$^{c}$±1.00 | 2.38$^{b}$±0.40 | 2.80$^{a}$±0.13 | 0.19 | 0.001 |
| Urea (mg/dl) | 5.38$^{b}$±0.12 | 4.07$^{b}$±0.24 | 7.60$^{a}$±2.40 | 4.82$^{b}$±0.3 | 7.25$^{a}$±0.42 | 0.44 | 0.001 |
| Uric acid (mg/dl) | 7.78$^{b}$±0.30 | 5.46$^{c}$±0.07 | 8.58$^{b}$±0.72 | 5.26$^{c}$±0.01 | 12.32$^{a}$±1.40 | 0.70 | 0.004 |
| Creatinine (mg/dl) | 0.20±0.06 | 0.18±0.02 | 0.18±0.02 | 0.24±0.01 | 0.30±0.01 | 0.01 | 0.775 |
| Calcium (mg/dl) | 9.98$^{b}$±0.81 | 6.20$^{c}$±0.88 | 10.11$^{a}$±0.9 | 10.56$^{a}$±1.01 | 7.42$^{c}$±0.65 | 0.50 | 0.003 |
| Phosphorus (mg/dl) | 3.60±0.52 | 3.35±0.90 | 4.07±0.20 | 4.05±0.07 | 4.04±0.33 | 0.14 | 0.339 |

1 = 22% CP + 2,900 kcal ME/kg feed; 2 = 22% CP + 2,600 kcal ME/kg feed; 3 = 24% CP + 2,600 kcal ME/kg feed; 4 = 26% CP + 2,600 kcal ME/kg feed; 5 = 28% CP+ 2,600 kcal ME/kg feed
$^{abc}$Means values within the same row with different superscripts differed significantly (P < 0.05).
g/dl = Gram per deciliter; mg/dl = Milligram per deciliter.

## Meat quality and color

The overall dressing percentage of broilers was slightly lower than the reference value because this study considered broilers without skin (Table 5). Breast meat yield was higher in diets containing higher protein levels in Groups 2, 3, 4, and 5, which had low energy levels. However, the thigh percentage was higher in Group 3, while the other groups had no consistencies for this parameter. The drumstick percentage was higher in Groups 3, 4, and 5 which were fed the high-protein and low-energy diets, respectively.

Birds fed low-energy and high-protein diets showed inconsistencies in meat crude protein, ether extract, dry matter, and L*, a*, and b* values ($P < 0.05$). The highest levels of crude protein (23.02%), dry matter (25.80%), a* (4.34), and b* (4.53) were found in Group 3, which was offered 24% CP and 2,600 kcal ME/kg. However, birds fed different levels of protein, but low energy (2,600 kcal/kg) exhibited substantially lower muscle fat (3.10%) and skin fat (3.05%) in Groups 3 and 4. The lowest levels of ether extract (0.23%) and lightness (L* 45.10) were observed in Groups 3 and 2, respectively.

**Table 5** Effect of different level of energy and protein on meat quality of broilers

| Parameters | Dietary groups (% CP + ME kcal/kg feed) | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|
| | 1 (22+2900) | 2 (22+2600) | 3 (24+2600) | 4 (26+2600) | 5 (28+2600) | | |
| **Carcass traits** | | | | | | | |
| Dressed weight (DW; % live weight) | 58.47$^{c}$±14 | 58.51$^{c}$±14 | 62.06$^{a}$±14 | 59.49$^{b}$±42 | 60.03$^{a}$±17 | 5.03 | 0.002 |
| Breast (% DW) | 38.84$^{c}$±11 | 42.03$^{b}$±2 | 47.22$^{a}$±5 | 40.00$^{b}$±11 | 40.99$^{c}$±8 | 1.95 | 0.001 |
| Thigh (% DW) | 30.43$^{b}$±14 | 25.82$^{c}$±5 | 34.44$^{a}$±11 | 28.19$^{b}$±16 | 28.71$^{b}$±25 | 3.50 | 0.004 |
| Drumstick (% DW) | 15.94$^{c}$±8 | 14.68$^{c}$±2 | 16.11$^{b}$±8 | 17.83$^{a}$±8 | 17.10$^{a}$±14 | 1.95 | 0.035 |
| Abdominal fat (% DW) | 3.62$^{b}$±7 | 3.16$^{c}$±7 | 3.12$^{c}$±3.0 | 3.91$^{a}$±3.0 | 4.10$^{a}$±3.0 | 1.10 | 0.003 |
| Skin fat (g) | 6.90$^{a}$±0.3 | 5.10$^{b}$±0.3 | 4.05$^{bc}$±0.2 | 3.05$^{c}$±0.3 | 5.25$^{b}$±0.3 | 0.35 | 0.002 |
| **Composition of meat (g /100 g fresh)** | | | | | | | |
| Dry matter | 24.40$^{b}$±2.2 | 23.94$^{c}$±2.34 | 25.80$^{a}$±2.12 | 24.59$^{b}$±2.3 | 25.40$^{a}$±1.87 | 0.51 | 0.004 |
| Crude protein | 21.98$^{c}$±2.2 | 22.54$^{b}$±1.9 | 23.02$^{a}$±2.12 | 22.78$^{b}$±1.6 | 22.76$^{b}$±1.52 | 0.42 | 0.001 |
| Ether extract | 2.48$^{a}$±0.21 | 0.83$^{c}$±0.33 | 0.23$^{c}$±0.15 | 1.29$^{b}$±0.12 | 2.96$^{a}$±0.24 | 0.28 | 0.002 |
| Ash | 1.10±0.11 | 1.26±0.08 | 1.34±0.31 | 1.31±0.11 | 1.27±0.09 | 0.04 | 0.338 |
| **Breast meat color** | | | | | | | |
| L* | 48.69$^{a}$±2.2 | 45.10$^{c}$±2.70 | 46.26$^{c}$±1.07 | 47.27$^{b}$±5.50 | 48.19$^{a}$±1.65 | 0.75 | 0.005 |
| a* | 3.02$^{c}$±1.20 | 3.56$^{b}$±1.10 | 4.34$^{a}$±0.07 | 3.28$^{c}$±0.46 | 3.20$^{c}$±1.06 | 0.23 | 0.004 |
| b* | 4.63$^{a}$±2.4 | 4.32$^{b}$±1.29 | 3.49$^{c}$±1.64 | 3.89$^{b}$±0.7 | 4.53$^{a}$±1.10 | 0.35 | 0.001 |

1 = 22% CP + 2,900 kcal ME/kg feed; 2 = 22% CP + 2,600 kcal ME/kg feed; 3 = 24% CP + 2,600 kcal ME/kg feed; 4 = 26% CP + 2,600 kcal ME/kg feed; 5 = 28% CP+ 2,600 kcal ME/kg feed.
$^{abc}$Means values within the same row with different superscripts differed significantly ($P < 0.05$).
*L = Lightness, *a = Redness, *b = Yellowness, DW = dressed weight without skin and giblet, g = Gram, % = Percentage

# DISCUSSION

## Performance

Increased protein levels enhanced muscle development, leading to better weight gain in Groups 3, 4, and 5 (Mahrose et al., 2015). In general, average weight gain in all groups was lower than expected, as indicated in the Cobb-500 management guide, due to using a mash diet as well as free from many growth-related additives, which allowed for the assessment of the real effect of the factors considered (Table 1). However, an increasing trend was not observed with increased protein, which might be due to the suitability of the protein-to-energy ratio found in Groups 3 and 4. However, Group 2 contained protein levels similar to Group 1, but lower energy was associated with increased feed consumption by the birds in Group 2, indicating that more protein intake also influenced weight gain (Mahrose et al., 2015). All lower-energy groups (Groups 2 to 5) consumed more feed because birds eat to satisfy their energy requirements (Classen, 2017). In addition, increased weight gain and better FCR (1.67) were observed in Group 3 (24% CP and 2,600 kcal ME/ kg diet) than in the other groups ($P < 0.05$). These results might be due to the higher amount of digested protein and its absorption, as well as the proper energy-to-protein ratio, because higher or lower doses of protein or energy suppress nutrient utilisation in broilers, resulting in poor growth performance (Marcu et al., 2013). In agreement with previous findings, it was observed in the current study that low-energy with high-protein (26% and 28%) resulted in lower body weight gain and worse FCR than the group containing 24% CP (Group 3). Moreover, birds fed 24% protein with low-energy showed better results and might retain more nitrogen in broiler muscles through optimal energy utilisation (Kamran et al., 2008). In the present study, high protein levels (26% and 28%) changed the energy-protein ratio (energy-to-protein ratio 100:1 and 93:1) and reduced the overall performance of birds which is similar to previous findings (Swatson et al., 2002).

## pH of different segments of the digestive tract

Classen et al. (2007), found that the pH values were 6.02 in the crop, 3.37 in the proventriculus, 3.27 in the gizzard, 6.40 in the duodenum, 6.50 in the jejunum, and 8.15 in the ileum that is similar to current findings ($P < 0.05$). High-protein content (24%) with low-energy may be the primary reason for the better pH in the digestive tract, as these optimal protein and energy ratios improve protein digestibility, leading to amino acid availability in the gut and resulting in lower pH in the duodenum. In addition, lower duodenum and colon pH were obtained in birds fed 24% protein with low-energy, indicating better availability of amino acids in the gut or a more favourable microbial environment (Rodjan et al., 2018). Amino acids have a pH of 5.0, which helps lower the pH of the gut, resulting in higher duodenal villi height and crypt depth, as well as improved nutrient absorption, especially nitrogen retention in broilers (Rodjan et al., 2018).

## Serum protein indices and minerals status

The higher calcium, albumin, urea, and uric acid levels may be due to the higher body weight of Group 3 (24% CP and 2,600 kcal ME/kg diet). This finding is consistent with that of Garcia (2000), who reported that a diet containing 24% protein increased muscle protein levels compared to a diet containing 17% protein.

## Meat quality and color

Carcass weight is 69–71% of the total weight in broilers, including the skin and neck (Fereidoun et al., 2007), where skin and visible fat comprise 8–20% of carcass weight (Dairo et al., 2010). In this study, the carcass weight was 58%, which is reasonable given the absence of skin and fat percentage. However, Group 3 found a higher dressing yield among the groups due to the best combination of protein and energy. In general, the breast and thigh percentages were higher than those found by other researchers, which were again the best in Group 3. Drumsticks increased with increased protein levels in Groups 3, 4, and 5, which also agreed with the above findings. Dietary levels of protein and energy in Group 3 were found to be suitable for lowering abdominal fat content. In general, lowering the energy content linearly decreased the fat content in the skin (Ferrini et al., 2008). The highest levels of crude protein (23.0%), dry matter (25.8%), leanness (0.23% fat), and a* (4.34) values were found in birds in Group 3 (24% protein group).

Meat color is one of the important quality parameters and showed significant variations between dietary groups. Group 3, with 24% crude protein and 2,600 kcal ME/kg feed, had the highest a* (4.34) and acceptable b* (3.49) value, hence the best quality meat (Rahman et al., 2022). Greater a* could be due to increased myoglobin content related to better protein-energy balance (Redoy et al., 2021). Conversely, lower L* values in Groups 2 and 3 suggest reduced oxidative stress and improved muscle integrity. These findings align with the preference for visually appealing, high-quality meat in the market, underscoring the importance of dietary formulations in influencing meat pigmentation (Tang et al., 2007).

# CONCLUSIONS

Under these circumstances, 24% crude protein and low-energy (2,600 kcal ME/kg feed) in the diet showed improved growth and better FCR, improved redness of meat, reduced fat content in meat, abdomen, and skin, as well as increased serum calcium levels compared with other combinations of protein and energy groups.

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# AUTHOR CONTRIBUTION

Mr. Abu Jafor Siddik his given his efforts and time to conduct the feeding trial data collection. Besides, data management, analysis and manuscript preparation have been done by Mr. Md. Aliar Rahman. The experiment was designed by Dr. Khan Md. Shaiful Islam and Mr. Md. Aliar Rahman. They supervised data collection and lab analysis. The manuscript write-up has been improved by Dr. Rakhi Chowdhury. All authors given their efforts to improve this manuscript.

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