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# Dietary Protein and Energy Levels at 3200 m Altitude for Slow-Growing Chickens

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**Abstract:** The extent to which energy and protein (CP) levels can be reduced without impacting productivity in slow-growing chickens remains undetermined. The treatments were conducted to evaluate the effect of dietary CP (17.5, 19, and 20.5%) and energy (2850-2950 kcal ME/kg) on slow-growing chickens using an ideal protein concept at a high altitude (3200 m). We used a total of 144 slow-growing chickens. The experiment lasted for eight weeks, a total of six treatments with three replicates each and eight chickens per replicate were used. Body Weight Gain (BWG) showed differences only in the growing chickens, for CP and the energy × CP interaction, the lowest BWG was at 17.5% CP (2850 kcal ME/kg) and 19.0% at 2950 kcal ME/kg ($p<0.01$) and higher for other interactions. BW and BWG maximized at 19% CP (2850 kcal ME/kg) and 17.5% CP (2950 kcal ME/kg). Feed intake was lower at 17.5 and 20.5% CP for respectively energy levels ($p<0.01$). Feed conversion was higher (finishing phase) with 20.5% CP (2850 kcal ME/kg) and 19% CP (2950 kcal ME/kg). Carcass yield was unaffected by treatment ($p>0.05$). Abdominal fat increased with high energy ($p<0.05$). At both energies, maturity weight was highest at the lowest CP; growth rate (kg/d) was raised with higher dietary CP (2850 kcal ME/kg). The time to maximize growth rate decreased with higher CP at low energy but remained constant at high energy. There is an interaction effect between energy and crude protein in the diet, where the highest response is with 19% PC for 2850 kcal ME/kg and 17.5% for 2950 kcal ME/kg.

**Keywords:** Energy, Protein Level, Feed Intake, Growth Rate, Slow-Growing Chicken, Ideal Protein Profile

## Introduction

Success in poultry production is based on effective feeding management and knowledge of the nutritional requirements, which vary depending on the genetic line, sex, age, and environmental conditions. To evaluate the carcass quality requires knowledge of the muscle mass and fat not used by the consumer (Deaton *et al.*, 1983). That's why it's necessary to know the energy and protein levels in the diet to maximize the deposition of muscle tissue and reduce fat in slow-growing chickens.

An adequate assessment of all these factors is necessary to design economic diets, even more knowing that currently, the variation in prices of the ingredients is highly fluctuating (Abiden *et al.*, 2019; Wu *et al.*, 2019). Among the nutritional components, energy, and protein are the ones with the greatest impact on the price of the diet (Cerrate *et al.*, 2019; Noblet *et al.*, 2022) and the production of farm animals (M. Abdel-Hafeez *et al.*, 2016). Therefore, the adequate use of protein and energy will influence the profitability of breeding.

Currently, there is a tendency to decrease dietary energy and protein in many countries, to reduce costs (Kamran *et al.*, 2008) and minimize nitrogen excretion (Hilliar *et al.*, 2020). In this context, it is important to reduce dependence on imported soybean meal (Greenhalgh *et al.*, 2022) and corn. However, this approach may have implications on protein utilization efficiency, leading to potential effects on productive performance, carcass quality (Abiden *et al.*, 2019; An and Kong, 2023; Musigwa *et al.*, 2020), and the environment due to inefficient use of nitrogen (Siegert *et al.*, 2023) and energy to metabolic level.

The relationship between energy and dietary protein is known. However, it has not been established to what extent it is possible to reduce protein and modify the energy without affecting the productive response of chickens (Musigwa *et al*., 2020). Considering this aspect, it has been proposed to formulate diets by manipulating dietary protein levels while maintaining the concept of ideal protein. However, these effects have been little studied in slow-growing chickens and the relationships between amino acids and energy levels have not been considered.

Studies carried out show that the reduction in dietary protein levels allows for significantly improved weight gain and feed conversion ratio (Greenhalgh *et al*., 2022). However, a series of experiments have demonstrated that reducing dietary crude protein in diets can negatively affect bird performance, leading to decreased growth rates and increased carcass fat content (Swennen *et al*., 2004). Based on these considerations, the challenge for a nutritionist is to formulate economically viable diets that provide amino acids and energy requirements as closely as possible for chickens (Nawaz *et al*., 2006) and enhance performance, while also reducing environmental pollution (Soomro *et al*., 2017).

Therefore, aimed to evaluate the effects of dietary protein (17.5, 19, and 20.5%) and energy (2850 and 2950 kcal ME/kg) in slow-growing chickens. The ideal protein concept was applied, which allows recommendations on dietary protein when energy levels are modified to efficiently utilize amino acids at the metabolic level.

## Materials Methods

### *Study Location and Duration*

The School of Zootechnics of the Universidad Nacional de San Antonio Abad del Cusco Peru conducted the study at 3219 m above sea level. The experiment lasted for eleven weeks where the chickens were reared together and fed a starter diet from the third weeks of age. The experimental evaluation had two phases: Grower from 21-49 days and finisher from 49-77 days of age.

### *Animal and Facilities*

One hundred and forty-four slow-growing chickens with an average weight of 424.33±0.06 gm were used. The chicken house covered an area of 51.60 m$^2$ (floor pen), partitioned into 18 pens (1.95×1.30 m), and equipped with hopper-type manual feeders and automatic drinkers. Temperature and humidity were recorded daily with a thermohygrometer (Isolab. Laborgerate GmbH. Germany) and to maintain a constant temperature, heaters were used. All chickens used in the study were vaccinated against Newcastle, Gumboro, and Bronchitis. Each experimental pen contained rice hulls as bedding at a depth of approximately 05 (five) cm. A total of six treatments with three replicates were randomized, considering 24 chickens for each treatment and eight chickens for each replication.

### *Experimental Diets*

Two energy levels (2850 and 2950 kcal/kg ME) and three dietary protein levels (17.5, 19, and 20.5%) were evaluated. Dietary protein was reduced by the ideal protein concept. Feeding was ad libitum (Tables 1-2).

**Table 1:** Diet composition for growing chicken (21-49 days) (As feed)

| Ingredients | 2850 kcal/kg | | | 2950 kcal/kg | | |
|---|---|---|---|---|---|---|
| | 17.5 % | 19.0 % | 20.5 % | 17.5 % | 19.0 % | 20.5 % |
| Corn | 59.04 | 55.88 | 60.83 | 63.37 | 60.23 | 57.76 |
| Soybean meal | 23.92 | 26.62 | 30.15 | 15.84 | 20.92 | 22.03 |
| Wheat by-products | 3.62 | 1.59 | 3.81 | 7.28 | 5.08 | 4.82 |
| Barley | 10.00 | 10.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| Full-fat Soybean | 0.11 | 2.74 | 0.93 | 10.00 | 10.00 | 10.00 |
| Fish meal | 0.00 | 0.00 | 1.07 | 0.00 | 0.00 | 2.00 |
| Soybean oil | 0.00 | 0.00 | 0.00 | 0.04 | 0.49 | 0.54 |
| Dicalcium phosphate | 0.89 | 0.86 | 0.73 | 0.97 | 0.94 | 0.67 |
| Calcium carbonate | 1.38 | 1.36 | 1.32 | 1.40 | 1.39 | 1.34 |
| Salt | 0.17 | 0.17 | 0.14 | 0.21 | 0.20 | 0.19 |
| DL-Methionine | 0.07 | 0.03 | 0.00 | 0.10 | 0.06 | 0.00 |
| L-Lysine | 0.06 | 0.00 | 0.21 | 0.10 | 0.00 | 0.00 |
| Sodium bicarbonate | 0.18 | 0.19 | 0.28 | 0.12 | 0.14 | 0.10 |
| Premix | 0.30 | 0.30 | 0.30 | 0.30 | 0.30 | 0.30 |
| Choline, 60% | 0.10 | 0.10 | 0.10 | 0.10 | 0.10 | 0.10 |
| Additives | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 |
| *Nutritional value, %* | | | | | | |
| Crude protein | 17.50 | 19.00 | 20.42 | 17.50 | 19.00 | 20.50 |
| Ether extract | 2.74 | 3.06 | 2.93 | 4.59 | 4.89 | 5.03 |
| Crude fiber | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 |
| Nitrogen-free extract | 60.39 | 58.45 | 57.08 | 58.33 | 56.47 | 54.86 |
| Metabolizable energy, kcal/kg | 2850 | 2850 | 2850 | 2950 | 2950 | 2950 |
| Lysine | 0.89 | 0.96 | 1.21 | 0.92 | 0.96 | 1.08 |
| Methionine | 0.36 | 0.34 | 0.34 | 0.39 | 0.37 | 0.35 |
| Methionine-Cysteine | 0.64 | 0.64 | 0.65 | 0.66 | 0.66 | 0.66 |
| Available phosphorus | 0.31 | 0.31 | 0.31 | 0.32 | 0.32 | 0.32 |
| Calcium | 0.80 | 0.80 | 0.80 | 0.83 | 0.83 | 0.83 |
| Sodium | 0.14 | 0.14 | 0.16 | 0.14 | 0.14 | 0.14 |

Premix: vitamins and minerals

**Table 2:** Diet composition for finisher chicken (49-77 days) (As feed)

| Ingredients | 2850 kcal/kg | | | 2950 kcal/kg | | |
|---|---|---|---|---|---|---|
| | 17.5 % | 19.0 % | 20.5 % | 17.5 % | 19.0 % | 20.5 % |
| Corn | 56.38 | 56.26 | 52.10 | 68.24 | 66.53 | 63.90 |
| Soybean meal | 18.57 | 24.12 | 27.65 | 15.11 | 22.24 | 27.67 |
| Wheat by-products | 6.78 | 3.93 | 1.34 | 8.23 | 3.25 | 0.00 |
| Wheat | 3.68 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| L-Threonine | 0.09 | 0.00 | 0.00 | 0.09 | 0.00 | 0.00 |
| Barley | 11.00 | 11.00 | 11.00 | 0.00 | 0.00 | 0.00 |
| Full-fat Soybean | 0.00 | 1.75 | 5.03 | 5.00 | 5.00 | 5.00 |
| Soybean oil | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.15 |
| Dicalcium phosphate | 1.29 | 1.22 | 1.18 | 1.28 | 1.25 | 1.22 |
| Calcium carbonate | 1.05 | 1.05 | 1.03 | 1.05 | 1.04 | 1.02 |
| Salt | 0.23 | 0.22 | 0.22 | 0.12 | 0.23 | 0.23 |
| DL-Methionine | 0.08 | 0.02 | 0.00 | 0.09 | 0.04 | 0.00 |
| L-Lysine | 0.14 | 0.00 | 0.00 | 0.15 | 0.00 | 0.36 |
| Sodium bicarbonate | 0.30 | 0.01 | 0.02 | 0.19 | 0.00 | 0.01 |
| Premix | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 |
| Choline, 60% | 0.08 | 0.08 | 0.08 | 0.08 | 0.08 | 0.08 |
| Additives | 0.20 | 0.20 | 0.20 | 0.20 | 0.20 | 0.20 |
| *Nutritional value, %* | | | | | | |
| Crude protein | 17.75 | 19.00 | 20.50 | 17.50 | 19.00 | 20.50 |
| Ether extract | 2.77 | 2.95 | 3.33 | 3.86 | 3.70 | 3.70 |
| Crude fiber | 4.00 | 4.00 | 4.00 | 3.90 | 3.72 | 3.62 |
| Nitrogen free extract | 61.51 | 59.50 | 56.95 | 61.04 | 59.42 | 57.50 |
| Metabolizable energy, kcal/kg | 2850 | 2850 | 2850 | 2950 | 2950 | 2950 |
| Lysine | 0.76 | 0.80 | 0.94 | 0.78 | 0.81 | 1.21 |
| Methionine | 0.31 | 0.29 | 0.29 | 0.34 | 0.31 | 0.30 |
| Methionine-Cysteine | 0.53 | 0.53 | 0.56 | 0.55 | 0.55 | 0.56 |
| Available phosphorus | 0.37 | 0.37 | 0.37 | 0.37 | 0.37 | 0.37 |
| Calcium | 0.75 | 0.75 | 0.75 | 0.75 | 0.75 | 0.75 |
| Sodium | 0.19 | 0.11 | 0.11 | 0.13 | 0.11 | 0.11 |

Premix: Vitamins and minerals

## Data Measurement

### *Productive Response*

Performance was assessed every seven days. At the end of the study, ten birds were randomly selected from each treatment to remain fasted and given only water (Moscoso-Muñoz *et al*., 2020). The chickens were weighed (Kern, PCB 6000) and euthanized (cervical dislocation) after 8 h of fasting (Caldas *et al*., 2019; Bandara *et al*., 2023), then bled and all feathers on the skin, legs, and viscera, including kidneys, were removed. The hot carcasses were chilled (water at 1ºC for four hours) and weighed to obtain the cold carcass weight (Ko *et al*., 2023). The abdominal fat pad was removed and weighed.

### *Laboratory Analysis*

Diets were analyzed following the guidelines of the AOAC (2005) Standardized Protocols for Animal Feed. Humidity using a forced convection oven (Binder, FED 720) at 135°C for 2 h (AOAC 930.15). Ash with a muffle (Protherm, ECO 119) at 600°C for 8 h (AOAC 942.05). Crude protein with an elemental analyzer CHNO/S (Perkin Elmer, 2400 Series II) (AOAC 990.03) and crude fat with an automatic Soxhlet extractor (Hanon, SOX 606) (AOAC 2003.05).

### *Evaluation of Growth Parameters*

The Gomperzt model was used to determine the growth parameters (Afrouziyeh *et al*., 2021). The first derivative was used to determine the weight (BW) and weight gain (BWG) for the average day: BW = A * e (-e (-B * (t-T))); BWG = Body BW*(Ln(A/BW)). Where: A: Weight at maturity, B: Growth rate, and T: Optimal time to maximize growth rate.

### *Statistical Analysis*

A Completely Random Design was used with a 2×3 factorial arrangement: Two energy densities (2850-2950 kcal) and three protein levels (17.5, 19, and 20.5%) with three replicates/pens (18 experimental units). All result was analyzed by ANOVA and a Tukey Post Hoc analysis was used to determine which group means differed at 5%. Before data analysis, normal distribution (Anderson and Darling test) and the homogeneity of variance (Levene test) were verified. To determine the percentage of error, the comparison was made between the values obtained and those estimated with the prediction model, first calculating the mean square of the predicted error and then the percentage of predicted error, expressed as follows (Nogueira *et al*., 2021):

$$MSPE = \sum_{i=1}^{n}(Oi - Pi)\hat{}2/n \quad \%\,\mathrm{e} = \sqrt{\mathrm{MSPE}} \times (100/\hat{y})$$

where, *MSPE*: is the mean square of the predicted error, *n*: is the total number of observations, *Oi*: is the observed value, *Pj*: is the predicted value, *ŷ*: is the average of the observed values, and % *e*: is the predicted error percentage.

## Results

### *Productive Response*

Both BW and BWG showed different responses. For the BW (growing and finished), only the energy × protein (CP) interaction showed significant differences ($p<0.01$).

The chickens fed 17.5% PC and 2850 Kcal ME had the lowest BW at the end of the growing phase. In the finishing phase, the treatments with 19% CP and 2850 Kcal ME and 17.5% CP with 2950 Kcal ME, had the highest BW (Fig. 1) and (Table 3).

BWG showed significant differences in the growth phase for CP and energy × CP interaction, where the lowest BWG was at 17.5% CP (2850 kcal ME) and 19.0% at 2950 kcal ME ($p<0.01$) and higher with other interactions. To finishing and total phase, no differences were observed among treatments (Table 3), but there was a tendency ($p = 0.06$) where the best response was with 17.5% CP (2950 kcal ME) and 19% CP with 2850 kcal ME to the finishing phase. The regression models showed that for 2850 kcal ME/kg, the BW and BWG were maximized at 19% CP (growth, finishing, and total). At 2950 kcal ME/kg, the best response was observed with 17.5% CP (Fig. 2).

Feed intake showed significant differences only for the energy x protein interaction in all feeding phases, which was low at 17.5-20.5% CP for 2850 and 2950 kcal ME/kg respectively ($p<0.01$). Feed conversion showed differences only in the finishing phase for the energy × CP interaction, with high feed conversion in the chickens fed 20.5% (2850 kcal ME/kg) and 19% (2950 kcal ME/kg) ($p<=0.05$) of CP.

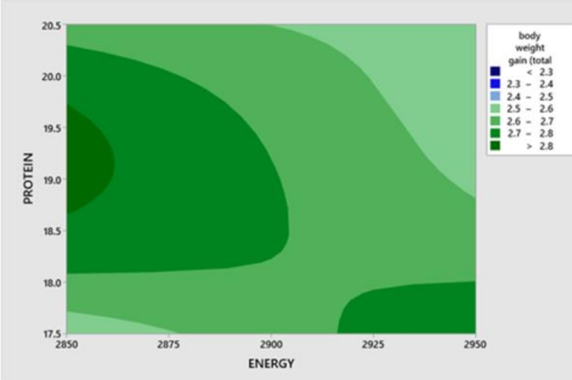


**Fig. 1:** Effect of energy and CP on body weight and body weight gain (total). L: Left; R: Right

**Table 3:** Productive response among treatments

| Variable | 2850 kcal/kg ME | | | 2950 kcal/kg ME | | | Energy | | Protein | | | P-value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 17.5 % | 19.0 % | 20.5 % | 17.5 % | 19.0 % | 20.5 % | 2850 | 2950 | 17.5 % | 19.0 % | 20.5 % | E | P | ExP |
| *Body weight, kg/bird* | | | | | | | | | | | | | | |
| Starter | 0.43 | 0.42 | 0.44 | 0.43 | 0.41 | 0.41 | 0.43 | 0.42 | 0.43 | 0.42 | 0.43 | 0.19 | 0.71 | 0.66 |
| Grower | $1.48^{b}$ | $1.69^{a}$ | $1.69^{a}$ | $1.66^{a}$ | $1.60^{a}$ | $1.62^{a}$ | 1.62 | 1.62 | 1.57 | 1.64 | 1.65 | 0.89 | 0.06 | 0.00 |
| Finisher | $2.96^{c}$ | $3.25^{a}$ | $3.09^{bc}$ | $3.22^{ab}$ | $2.99^{bc}$ | $2.97^{bc}$ | 3.10 | 3.06 | 3.09 | 3.12 | 3.03 | 0.57 | 0.56 | 0.01 |
| *Body weight gain, kg/bird* | | | | | | | | | | | | | | |
| Grower | $1.05^{b}$ | $1.27^{a}$ | $1.25^{a}$ | $1.23^{a}$ | $1.18^{b}$ | $1.20^{a}$ | 1.19 | 1.21 | $1.14^{b}$ | $1.22^{a}$ | $1.23^{a}$ | 0.56 | 0.04 | 0.00 |
| Finisher | 1.08 | 1.14 | 0.95 | 1.13 | 1.05 | 0.97 | 1.06 | 1.04 | 1.10 | 1.07 | 0.96 | 0.69 | 0.07 | 0.28 |
| Overall | 2.53 | 2.83 | 2.65 | 2.79 | 2.58 | 2.55 | 2.67 | 2.64 | 2.66 | 2.71 | 2.60 | 0.72 | 0.52 | 0.06 |
| *Feed intake, kg/bird* | | | | | | | | | | | | | | |
| Grower | $4.93^{b}$ | $5.48^{a}$ | $5.45^{a}$ | $5.50^{a}$ | $5.38^{a}$ | $4.96^{b}$ | 5.29 | 5.28 | 5.22 | 5.43 | 5.20 | 0.97 | 0.15 | 0.00 |
| Finisher | $3.19^{ab}$ | $3.63^{a}$ | $3.75^{a}$ | $3.44^{ab}$ | $3.60^{a}$ | $2.93^{b}$ | 3.52 | 3.32 | 3.32 | 3.61 | 3.34 | 0.22 | 0.26 | 0.04 |
| Overall | $8.12^{bc}$ | $9.11^{a}$ | $9.20^{a}$ | $8.95^{ab}$ | $8.98^{ab}$ | $7.89^{c}$ | 8.81 | 8.61 | 8.53 | 9.04 | 8.55 | 0.40 | 0.16 | 0.01 |
| *Feed: gain, kg/kg* | | | | | | | | | | | | | | |
| Grower | 4.71 | 4.33 | 4.35 | 4.47 | 4.58 | 4.15 | 4.46 | 4.40 | 4.59 | 4.45 | 4.25 | 0.61 | 0.13 | 0.26 |
| Finisher | $2.98^{b}$ | $3.18^{b}$ | $3.95^{a}$ | $3.04^{b}$ | $3.62^{a}$ | $3.07^{b}$ | 3.37 | 3.25 | 3.01 | 3.40 | 3.51 | 0.50 | 0.09 | 0.03 |
| Overall | 3.22 | 3.23 | 3.47 | 3.21 | 3.50 | 3.10 | 3.31 | 3.27 | 3.21 | 3.36 | 3.29 | 0.76 | 0.56 | 0.10 |

Different letters in the same line differ significantly ($p<0.05$). Where: E: Level of energy, P: Level of protein, E × P: Energy × protein interaction

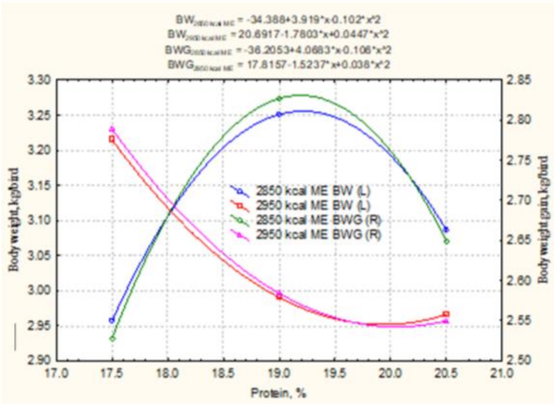


**Fig. 2:** Effect of treatments on body weight and BWG (total). L: Left; R: Right

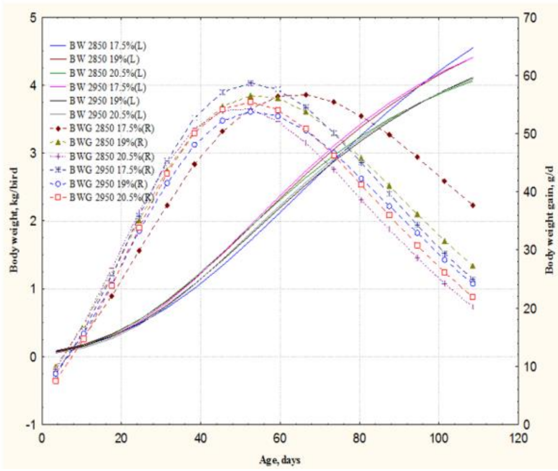


**Fig. 3:** Body weight and body weight gain (Gompertz model). L: Left; R: Right

*Yield of Carcass and Abdominal Fat*

Carcass weight and percentage yield were not affected by variations in energy and protein diets. However, a positive trend was observed between protein percentage and the carcass yield ($R^2$ 0.99), especially at the highest energy level. When evaluating carcass weight (kg), the positive trend occurred with the lowest energy ($R^2$ 0.98). Abdominal fat weight showed differences between energy levels ($p<=0.054$), being higher with 2950 kcal/kg ME than other treatments. No effect was observed on CP or energy × CP interaction. On the other hand, there were no differences when abdominal fat was expressed as a percentage of the carcass. However, a positive trend was observed between CP and abdominal fat (%), with high energy in the diet ($R^2$ 0.93), where the higher the protein content in the diet, the greater the percentage of fat deposition (Table 4).

## Growth Parameters

The growth parameters (weight at maturity, growth rate, and time to maximum growth rate), showed that, at both energy levels, the weight at maturity was highest with the lowest dietary CP (17.5%) and tended to decrease with the increase in CP. The growth rate (kg/d) showed a different behavior between energy levels, at 2850 kcal ME/kg, which increased with the increase in dietary CP. At the highest energy level, the growth rate remained constant among CP. The time to maximize growth rate decreased with increasing dietary CP (lower energy), but at high energy, this decrease was not marked and remained constant (Fig. 3) and (Table 5).

Figure (3) Body weight and body weight gain (Gompertz model). L: Left; R: Right.

.

**Table 4: Effect of variation in energy and protein on carcass yield and abdominal fat**

| Variables | 2850 kcal/kg ME | | | 2950 kcal/kg ME | | | Energy | | Protein | | | P-value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 17.5 % | 19.0 % | 20.5 % | 17.5 % | 19.0 % | 20.5 % | 2850 | 2950 | 17.5 % | 19.0 % | 20.5 % | E | P | Ex P |
| Body weight, kg | 3.08 | 3.19 | 3.19 | 3.35 | 2.99 | 3.06 | 3.15 | 3.13 | 3.22 | 3.13 | 3.09 | 0.85 | 0.62 | 0.15 |
| Carcass weight, kg | 2.44 | 2.53 | 2.58 | 2.61 | 2.39 | 2.45 | 2.52 | 2.48 | 2.53 | 2.46 | 2.52 | 0.72 | 0.81 | 0.30 |
| Yield of carcass, % | 79.62 | 79.22 | 80.79 | 77.92 | 79.88 | 80.21 | 79.88 | 79.33 | 78.77 | 79.55 | 80.50 | 0.67 | 0.54 | 0.75 |
| Abdominal fat, kg | 0.07 | 0.06 | 0.07 | 0.08 | 0.08 | 0.09 | 0.07$^b$ | 0.09$^a$ | 0.08 | 0.07 | 0.08 | 0.04 | 0.20 | 0.17 |
| Abdominal fat, % | 3.54 | 2.29 | 2.96 | 3.19 | 3.57 | 3.71 | 2.93 | 3.49 | 3.36 | 2.93 | 3.33 | 0.06 | 0.38 | 0.07 |

Different letters in the same line differ significantly ($p<0.05$)

**Table 5:** Growth parameters (Gompertz) in the treatments

| Parameter | 2850 kcal/kg ME | | | 2950 kcal/kg ME | | |
|---|---|---|---|---|---|---|
| | 17.5% | 19.0% | 20.5% | 17.5% | 19.0% | 20.5% |
| A, kg | 6.42 | 5.50 | 4.78 | 5.32 | 5.04 | 4.86 |
| B, kg/day | 0.024 | 0.028 | 0.031 | 0.030 | 0.029 | 0.031 |
| T, days | 64.20 | 54.51 | 49.61 | 52.88 | 53.59 | 51.96 |
| Error, % | 2.20 | 2.01 | 1.60 | 2.91 | 1.10 | 2.13 |

## Discussion

This study shows how energy and protein intake interact. When protein intake exceeds the body's requirements, the excess nitrogen is catabolized, resulting in increased urinary nitrogen excretion (Swennen *et al*., 2004). This process generates substantial heat (as protein has the highest heat increment among nutrients) (De Faria *et al*., 2007), which must be dissipated to the environment (Oliveira *et al*., 2013). Conversely, when dietary protein intake is low, it fails to meet the requirements of animals and hinders the synthesis of certain non-essential amino acids, including glycine, which is crucial for uric acid synthesis (van Milgen, 2021). This deficiency can lead to increased heat production by elevating serum levels of the thyroid-derived hormone triiodothyronine, known for its calorigenic effect (De Faria *et al*., 2007). In both scenarios where protein is either in excess or deficient, energy utilization and productive performance are adversely affected.

It should be noted that the productive response depends not only on the levels of amino acids in the diet but also on the feed intake, which is mainly related to the energy concentration of the diet (Hu *et al*., 2021). Reducing dietary protein (low levels) can produce depressed apparent ileal digestibility of amino acids (Greenhalgh *et al*., 2022). Blood protein and ammonia levels decrease, resulting in an imbalance of amino acids supply relative to theoretical requirements (alteration of the plasma amino acid pattern) and adverse effects due to the amino acids interactions (Cabezas-Garcia *et al*., 2022), which affects animal appetite, alters consumption by modulating the synthesis of neurotransmitter metabolites in the brain (Kamran *et al*., 2008), depressed weight gain, compromised feed conversion and increased relative abdominal fat pad weights (Greenhalgh *et al*., 2022).

However, when the reduction is moderate, the protein efficiency ratio increases (Kamran *et al*., 2008), and chicks fed high ME diets with normal energy/protein ratio grow faster and use feed more efficiently (M. Abdel-Hafeez *et al*., 2016). This would have been the case in the present study since the best productive response was obtained with 19% crude protein in the diet with the lowest energy level and 17.5% CP in the diet with the highest energy level, implying a more efficient use of N in broilers reared on diets with low CP (Musigwa *et al*., 2020) than in diets with low energy levels: CP ratio, where the productive response was low.

Various studies reported that dietary CP did not affect the growth performance of broilers (Kamran *et al*., 2008) and that the reduction of CP modulates the composition of the fecal microbiota (Laudadio *et al*., 2012a) contributes to the reduction of nutrient excretion, especially nitrogen (Laudadio *et al*., 2012b) and increases the protein efficiency ratio with low CP diets during the growth period (M. Abdel-Hafeez *et al*., 2016; Musigwa *et al*., 2020). On the other hand, some studies showed that there was an effect, as observed in laying hens, where the reduction in CP resulted in irregular clutch patterns (Cabezas-Garcia *et al*., 2022) and impaired performance (Oliveira *et al*., 2013). However, Laudadio *et al*. (2012b) reported that mean final BW tended to increase with increasing dietary protein and was significantly higher in the average protein group than in the high or low-protein diet.

These contradictions would be determined by the degree of reduction in dietary protein and interactions with energy, which was not the same in all the studies. A similar result was observed in the present study because there was a correlation between dietary CP and ME and, therefore, hence the optimal value for one cannot be predicted without considering the other (Zaman *et al*., 2008).

In this study, a positive trend was observed between protein percentage and carcass yield (%), mainly at the highest energy level. However, when carcass weight (kg) was considered, the positive trend occurred at the lowest energy level, implying that increasing dietary protein determines the highest carcass yield (Zaman *et al*., 2008). Otherwise, the abdominal fat weight was high at the highest energy level, which would be because an excess of energy relative to protein intake results in significant heat production and energy retention as fat (Musigwa *et al*., 2020). These results could be due to the interaction or relationship that exists between energy and protein, which determines variations in the efficiency of nutrient utilization (Swennen *et al*., 2004). The same effect is observed when the energy density is increased in the diet of chickens (Arjona and Guevara, 2019).

In support of these findings, a study (M. Abdel-Hafeez *et al*., 2016), showed that increasing dietary protein and ME levels significantly increased BWG, abdominal fat and liver weights, and protein and fat content of the carcass. Breast weight and fat content of the meat increased linearly with increasing CP of the diets, but also the low CP diet and reduction of dietary CP during rearing increases abdominal fat pad.

When analyzing the growth parameters, they were influenced by the variations in energy and dietary protein. It is observed that the weight at maturity is highest with the lowest level of protein at both energy levels in the diet and tends to decrease with the increase in protein (inverse relationship). These models confirm the results found in the present study, where the interaction between energy and protein, even with the ideal protein, shows a differentiated response between treatments. Reducing dietary protein levels can significantly improve weight gain and feed conversion ratio (Greenhalgh *et al*., 2022). Zaman *et al*. (2008) reported that high CP with low ME reduces BWG, probably because less energy is available to excrete metabolites of protein catabolism. In addition, the efficiency with which dietary protein is used affects animal nitrogen excretion and the environmental impact of animal production (van Milgen, 2021). Using the ideal protein concept, it is possible to reduce dietary protein to minimize the negative effects caused by excess nitrogen at the metabolic level and measure the impact of the inclusion of protein sources in the diet of chickens on the level of their excretion (Cabezas-Garcia *et al.*, 2022).

## Conclusion

There was an interactive effect between energy and crude protein in the diet, where the greatest response was with 19.5% protein for 2850 kcal ME/kg and 17% for 2950 kcal ME/kg. These results show that the reduction of dietary protein by applying the ideal protein allows for improving the productive response of chickens and minimizing nitrogen excretion, thereby maximizing economic and environmental benefits. Considering the existing interaction between energy and protein levels, it can be suggested that the reduction of dietary protein with increased energy levels should be considered for slow-growing chickens.

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## Author's Contributions

**Mariela Ketty Meza-Morveli:** Methodology, investigation, resources, project administration, funding acquisition.

**Liz Beatriz Chino-Velasquez:** Conceptualization, methodology, investigation, writing-original draft preparation, writing review and editing.

**Jesús Camero-DeLaCuba:** Methodology, supervision, project administration.

**Mario Arjona-Smith:** Methodology, data curation, writing-original draft preparation, supervision.

**Oscar Gomez-Quispe:** Conceptualization, data curation, writing-original draft preparation, writing-review and editing.

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## Ethics

All experimental procedures were approved by the Research Projects Approval Commission of the Professional School of Zootechnics, Faculty Agronomy of Zootechnics – UNSAAC (R-D-1276-2019-FCA).

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