



Adjustment of Metabolizable Energy Concentration in Diets for High-Laying Hens in the Subhumid Tropics

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Abstract

Egg production in hens in the subhumid tropics is low due to heat stress; therefore, the energy concentration of the diet must be adjusted to meet this need. The objective of this study was to evaluate different metabolizable energy concentrations in diets for hens. Eighty-four 20-week-old females with an initial weight of $1,595.3 \pm 9.9$ g were used. Different metabolizable energy concentrations (kcal/kg of feed) were evaluated: (ME_{2617} , ME_{3300} , ME_{3470} and ME_{3500}). Hen and egg production variables were measured using a completely randomized experimental design with weight as a covariate and a Tukey test. The laying percentage and feed conversion were similar ($p > 0.05$). Feed and amino acid consumption were higher ($p < 0.05$) in the ME_{2617} feed compared to the ME_{3500} feed. Average egg weight and width were greater ($p < 0.05$) in diets containing ME_{2617} , ME_{3300} and ME_{3470} compared to ME_{2617} (49.5 g and 40.5 mm). It is concluded that a higher concentration of metabolizable energy in the diet of hens in the subhumid tropics improves egg characteristics, decreases feed intake and decreases heat stress.

Keywords: Productivity; Production; Consumption; Poultry; Layers

Introduction

In the tropics, heat stress is the most important factor affecting the performance of high-laying hens, affecting feed intake, reproductive performance, posture, well-being, the endocrine system, acid-base imbalance, and organ functions [1]. Productive performance in Brown Nick hens is primarily due to genetic specialization and thermal comfort, producing over 300 eggs per hen [2,3]; however, this strain of birds is not suitable for all climates [2]. Brown Nick hens do not tolerate temperatures above 30°C [3]. Therefore, productive performance and diet should be studied in warm, subhumid climates.

Egg production is affected by ambient temperature, feed intake, and the energy density of the diet [3]. The optimal comfort temperature for hens is between 21 and 25°C, with some fluctuations up to 28°C [4-6]. As temperatures increase from 28 to 35°C, feed intake decreases by up to 36% due to heat stress [6] and growth rate and egg production also decrease [4].

In warm climates, the limiting factor for production is the bird's ability to avoid heat stress [6]. Hens maintained at temperatures of 27 to 33°C for 20 weeks show a decrease in feed intake [7], and consequently, a decrease in egg production.

A diet deficient in metabolizable energy (ME) causes birds to consume greater amounts of feed; On the contrary, if the energy content is higher, the consumption will be lower [8]. Birds fed with commercial feed in the subhumid tropics show an increase in feed consumption [9]. This indicates deficiencies in the amount of ME because the animals eat mainly to satisfy their energy needs [6,10]. The energy density of the recommended diet is 2684 to 2,992 kcal of ME/kg of feed [10].

However, feed consumption of birds in warm subhumid climates increases with these energy values [9]. On the other hand, it is mentioned that the percentage of laying (91.7%) is not affected in hens with different levels of ME (2,519 to 3,078 Kcal/kg) [11,12]. However, contradictions have been recorded [13]. Diets with 2,700 kcal ME and 15% crude protein increase feed intake beyond the recommended intake in the comfort zone [14,15], resulting in hens with high abdominal fat content.

In warm, subhumid climates, hens' feed intake is altered, resulting in decreased productivity. Therefore, it is necessary to adjust the amount of metabolizable energy and nutrients offered to hens to meet their needs [14]. Using lipids increases the energy density of the diet but dilutes the protein concentration [16].

Therefore, this study was conducted to test the hypothesis that the production of Brown Nick hens will be improved by adjusting the concentration of metabolizable energy in the feed. The objective of this study was to evaluate the effect of adjusting the metabolizable energy concentration of feed on the productive efficiency of high-laying Brown Nick hens raised in warm, subhumid climate conditions.

Materials and Methods

Geographic Location of the Study Area

The research was conducted in a Egg-laying hen farm in Bajos de Chila, Municipality of San Pedro Mixtepec, District 22. The geographic coordinates corresponding to 15°54'40.47" N, 97°7'58.37" W, with an altitude of 13 m above sea level [17], climate categorized as warm subhumid, with average annual rainfall of 500 to 2,500 mm and precipitation of the driest month between 0 and 60 mm; summer rainfall between 5 and 10.2% annually [18].

In the housing area, the maximum and minimum temperature were 32.5 and 27.7 °C respectively, with an average of 29.9 °C. The maximum and minimum relative humidity were 81.2 and 63.9% respectively, with an average of 72.4% (KT) thermohygrometer, model TA318).

The comfort of the females was ensured so that they did not suffer from stress at any time. However, the only exception was the heat stress by the predominant climate of the study area. This study was conducted on a family farm in the community of Bajos de Chila [9]. This study was endorsed by the Universidad del Mar, Puerto Escondido Campus in the official document: No. JCZPE/117/2021. The National Health Service, food safety, and quality was also taken into account.

Breeding and Selection

High-laying Brown Nick pullets were raised on the barn floor from day one of birth, in accordance with established management [19]. At the beginning of the laying period, 84 hens with an average weight of $1,595.3 \pm 9.9$ g and 20 weeks of age were selected. The hens were housed in cages for 15 days to become acclimatized to handling and feed consumption, followed by a three-month evaluation period.

Housing, Cages, Waterers and Feeders

The housing was 5 x 5 x 2.5 m in length, width, and height, respectively, with a sheet metal roof, chicken wire walls (to prevent the entry of rodents and birds), and a cement floor with a 3% slope. The cages used were commercially available, measuring 1.5 m in length with four compartments measuring 37 x 45 x 45 cm in width, depth, and height, respectively [9]. Each division had an individual tray for collecting feces. An automatic hummingbird-type drinker and a 250-g feeder were used in each division. At the beginning of the experiment, the barn was cleaned, scrubbed, and limed to keep the area as hygienic as possible. Feeders and waterers were cleaned daily.

Dietary Treatments

Four diets formulated with different levels of metabolizable energy (ME kcal/kg feed) were evaluated: ME₂₆₁₇, ME₃₃₀₀, ME₃₄₇₀, and ME₃₅₀₀. We began with the assumption that in warm subhumid climates, the ME requirement increases above 3,000 kcal/kg for egg production [6,12].

The diets were formulated with the Solver program (software used to formulate rations with a linear progression), according to two criteria: the nutritional needs of high-laying hens (Table 1) [14,20,21], and the nutrient intake of commercial diets. The change in diet from wheat bran to sorghum is due to the fact that the former provides lower levels of both metabolizable energy and lysine compared to sorghum. The amount of metabolizable energy, amino acids, and minerals in the diets were estimated according to Santiago, et al. [15] with support from tables and proximate chemical analysis [15,22].

Ingredient	Energy concentration of diets prepared on the basis of metabolizable energy (ME kcal/kg of food)			
	ME ₂₆₁₇	ME ₃₃₀₀	ME ₃₄₇₀	ME ₃₅₀₀
Soy	25.25	27.7	28.76	28.8
Corn	38.71	43.86	10	10
Wheat Bran	16.22	----	----	----
Sorghum	----	2	30.88	30.3
lysine	0.04	----	----	----
Methionine	0.16	0.14	0.14	0.14
Molasses, sugarcane	9.56	----	----	----
Oil	----	6.57	10.59	11.01
Commercial Vitamins	0.1	0.1	0.1	0.1
Phosphate rock	0.1	1.89	1.86	1.9
Calcium carbonate	1.86	7.42	7.42	7.41
Canthaxanthin	7.94	0.006	0.006	0.006
Salt	0.006	0.31	0.31	0.31
Calculated nutrient composition of diets				
Crude Protein	15.47	17.09	17.09	17.09
Metabolizable energy	2617	3300	3470	3500
Lysine	0.8	0.8	0.8	0.8
Methionine	0.4	0.4	0.4	0.4
Calcium	3.75	3.75	3.75	3.75
Phosphorus	0.55	0.55	0.55	0.55

Table 1: Percentage composition and nutrients of feed prepared for high-laying Brown Nick hens in subhumid tropics.

Management

Food and water were offered ad libitum every day. The living space provided a total of 16 hours of artificial and natural light daily [9,14]. A Fulgor TA318 timer was installed to automatically turn on the light. To reduce the temperature of the warehouse, ventilation was provided with a plastic fan (Man brand, high speed, air displacement of 105 m³/min and vertical rotation of 330°), which was kept on from 8:00 a.m. to 8:00 p.m. each day [9]. The birds were on a pre-established vaccination schedule [14,19] Variables evaluated.

Final weight (g), weight gain or loss (g), and feed intake (g) were assessed using an electronic scale (Rhino brand, model Bapo-10 with a capacity of 10 kg and an accuracy of 1g. Daily intake of metabolizable energy (kcal), crude protein (g), methionine (g), lysine (g), calcium (g), and phosphorus (g) were estimated [15,22]. The true feed conversion ratio was obtained by calculating feed intake (g) by the egg mass produced plus the hens' weight gain or loss. The laying

percentage was determined using Rojas' formula. The laying percentage was determined by means of the Rojas formula [23]. The variables of length, width and thickness of the eggshell were evaluated for three consecutive days each month using a digital micrometer (Sure bilt brand, Caliper Black model) [24]. Egg weight was measured using a digital scale (Pocket Scale, model 10053, approx. 0.01 g). The morphological, yolk, and white indices were determined using a depth bar with a vernier caliper (Sure Bilt brand, Caliper Black model) [25].

Statistical Analysis

A completely randomized experimental design was used for the analysis of the variables, with the initial weight of the breeding hens as a covariate. Four treatments and seven replicates were used, each experimental unit consisting of three hens. When differences were found, Tukey's mean separation test was used [26].

Results

Productive Efficiency of Brown Nick Hens

Treatments with different metabolizable energy contents behaved similarly ($p > 0.05$) regarding the variables of feed

conversion (3.1) and laying percentage (57.1%) (Table 2). Hens fed ME₂₆₁₇ showed an increase ($p < 0.0001$) in daily (18.0 g/bird) and final (2,077 g) weight, while treatments with higher ME content had weight losses of 1.5 g/bird/d and low final weight.

Diet	Weight (g) Final	Daily Weight Loss or Gain	Feed conversion	Laying (%)
ME ₂₆₁₇	2,076.6a	17.95a	3.1	59.3
ME ₃₃₀₀	1,455.3b	-1.65b	2.9	62.9
ME ₃₄₇₀	1,477.6b	-1.41b	3.3	57.7
ME ₃₅₀₀	1,479.1b	-1.34b	3.1	48.7
Average	1,622.10	3.4	3.1	57.1
F-value	23.19	36.27	0.14	1.08
SEM1	58.9	1.78	0.17	2.76
Probability	<0.0001	<0.0001	0.933	0.3835

^{a, b} Different superscripts in the same column indicate difference ($p < 0.0001$). ¹SEM = Standard Error of the Mean.

Table 2: Productive response in Brown Nick hens fed with different levels of metabolizable energy: ME₂₆₁₇, ME₃₃₀₀, ME₃₄₇₀ y ME₃₅₀₀ kcal/kg of feed, in subhumid tropics (n = 84).

Brown Nick hens fed ME₂₆₁₇ showed 32% higher feed intake ($p < 0.05$) and higher intake of the amino acids methionine, lysine, calcium, and phosphorus (g) compared to those fed ME₃₅₀₀ (Table 3). However, there were no

differences ($p > 0.05$) in protein and metabolizable energy intake per day, with average values of 16.5 g and 315.6 kcal/d/bird, respectively.

Diet	Daily Consumption (g) Feed	Protein	Methionine	Lysine	Calcium	Phosphorus	ME (Kcal)/d
ME ₂₆₁₇	123.1a	19.05	0.49a	0.99a	4.62a	0.68a	322.3
ME ₃₃₀₀	93.2ab	15.92	0.37ab	0.74ab	3.49ab	0.51ab	307.4
ME ₃₄₇₀	97.9ab	16.74	0.39ab	0.78ab	3.67ab	0.54b	339.8
ME ₃₅₀₀	83.7b	14.3	0.33b	0.67b	3.14b	0.46b	292.8
Average	99.47	16.5	0.4	0.8	3.73	0.55	315.6
F-value	4.76	2.39	4.77	4.77	4.46	4.75	0.67
SEM1	4.64	0.69	0.19	0.37	0.14	0.14	12.11
Probability	0.013	0.1025	0.0129	0.0129	0.0129	0.013	0.5806

^{a, b} Different superscripts in the same column indicate difference ($p < 0.0500$).

¹SEM = Standard Error of the Mean.

Table 3: Daily feed intake/Brown Nick hens fed different levels of metabolizable energy: ME₂₆₁₇, ME₃₃₀₀, ME₃₄₇₀ y ME₃₅₀₀ kcal/kg feed, in subhumid tropics (n = 84).

Egg Quality Assessment

The dietary treatments (ME₃₃₀₀, ME₃₄₅₀ y ME₃₅₀₀) produced 13.9% heavier eggs ($p < 0.01$) and 6.1% wider eggs ($p < 0.05$) compared to treatment ME₂₆₁₇. There were no

differences ($p > 0.05$) between the treatments evaluated for egg length, with an average value of 54.3 mm; however, shell thickness was 5.7% thicker in treatments ME₃₃₀₀ and ME₃₅₀₀ compared to ME₂₆₁₇ (Table 4).

Diet	Egg Weight (g)	Morphology (mm) Length	Width	Shell Thickness
ME ₂₆₁₇	49.54b	53.62	40.47b	0.33b
ME ₃₃₀₀	57.34a	54.46	42.96a	0.35a
ME ₃₄₇₀	58.02a	54.53	43.25a	0.32ab
ME ₃₅₀₀	57.21a	54.66	43.15a	0.35a
Average	55.53	54.31	42.45	0.33
F-value	11.8	0.7	20.47	7.69
SEM1	0.9	0.29	0.27	0.006
Probability	0.0002	0.5665	< 0.0001	0.016

^{a, b} Different superscripts in the same column indicate difference ($p < 0.05$).

¹SEM = Standard Error of the Mean.

Table 4: Egg weight and structure of Brown Nick hens fed with different levels of metabolizable energy: ME₂₆₁₇, ME₃₃₀₀, ME₃₄₇₀ y ME₃₅₀₀ kcal/kg of feed, in subhumid tropics (n = 84).

The morphological index for feeds containing ME₃₃₀₀, ME₃₄₇₀ y ME₃₅₀₀ was 4.5% higher ($p < 0.01$) compared to the ME₂₆₁₇ treatment (Table 5). However, the egg white index was

20% higher ($p < 0.05$) for the ME₂₆₁₇ treatment compared to the ME₃₄₇₀ treatment. The yolk index was similar ($p > 0.05$) across all four treatments.

Diet	Index Morphological	Yolk	White
ME ₂₆₁₇	75.52b	48.1	13.81a
ME ₃₃₀₀	78.99a	47.29	12.59ab
ME ₃₄₇₀	79.39a	48.7	11.00b
ME ₃₅₀₀	78.96a	47.65	12.04ab
Average	78.21	47.94	12.36
F-value	6.78	0.46	3.19
SEM1	0.43	0.43	0.38
Probability	0.003	0.716	0.0486

^{a, b} Different superscripts in the same column indicate difference ($p < 0.05$). ¹SEM = Standard Error of the Mean.

Table 5: Egg indices of Brown Nick hens with different levels of metabolizable energy: ME₂₆₁₇, ME₃₃₀₀, ME₃₄₇₀ y ME₃₅₀₀ kcal/kg of feed, in subhumid tropics (n = 84).

Discussion

Productive Efficiency in Brown Nick Hens

High-laying hens by 35 weeks of age should weigh 1,940g [14]. The lack of weight gain in hens fed ME₃₃₀₀, ME₃₄₇₀ and ME₃₅₀₀ could be due to six factors: 1) The nutrients in the diets remained the same amount per kilogram of feed, since only energy was concentrated; therefore, the hens did not meet the requirements of essential amino acids and minerals, making use of body reserves [5,8,10,27]. 2) Heat stress, house temperature and humidity were high; Brown Nick hens must be at optimal temperature of 18 to 24 °C and relative humidity of 50 to 60% for production; They do not tolerate temperatures above 30 °C [3,6,14]. This is why the hens showed panting to eliminate the excess heat produced, since they do not have sweat glands to withstand extreme

temperatures for a long time [3,6,28,29]. 3) Maintenance, in heat stress a high amount of metabolizable energy is required for the maintenance of homeostasis, as well as for egg and muscle production [6,30,31]. 4) Modern poultry genotypes have greater metabolic activity and produce more body heat [33]. However, higher egg production, larger eggs and lower feed consumption are expected [32]. 5) Farming system, the hens were kept on a roof with poor thermal insulation in hot weather, which could result in radiant heat gain on the birds' backs [33]. Additionally, conductive heat loss with cages is unlikely because of the small contact area with the hens [34]. 6) Hens kept at temperatures of 27 to 33 °C for 20 weeks decrease feed consumption [7].

The ambient temperature influenced the decrease in the birds' feed consumption [9], weight gain and egg production

[31,32]. The birds in this study were seen at the drinkers, panting, motionless, raising their wings [32]. However, the higher feed consumption in birds with ME₂₆₁₇ suggests that the amount of metabolizable energy provided by the diet is below the need in hens subjected to heat stress [14]. While with the other diets the level of metabolizable energy was adequate, since the birds obtained lower daily consumption [35,36]. When hens consume greater amounts of feed, they have the tendency to synthesize more muscle mass and body fat [37].

Genetic selection in high-laying hens has achieved the production of larger eggs with lower feed consumption with values of 2.03 to 2.07 [14,38], being higher in the present study. In a warm subhumid climate using diets with low energy levels ME₂₆₁₇, it has been recorded that feed consumption increases, in the temperature range of 23.3 to 32.8 °C and relative humidity of 52 to 64% [9]. However, when temperature and humidity increase, as in the present study, feed consumption decreases; therefore, also of amino acids, which causes lower egg production [12,39]; so it is necessary to increase the energy level of the diet and adjust the level of other nutrients.

Egg production and weight were not affected by the energy content of the diets [12,36,40], which was consistent in the present study. However, regarding hens consuming feed containing ME₂₆₁₇, the eggs were smaller in size relative to diets higher in metabolizable energy. The size of pullet eggs in Brown Nick hens in warm subhumid climate conditions are classified as medium [14,15,38,41].

Despite heat stress, the hens produced eggs with good shell thickness and are considered suitable for commercialization [42-45]. Heat stress did not delay follicular development and ovulation in hens, indicating that hormonal functions were normal in the ovarian follicles, and there was no decrease of prolactin, gonadotropins (follicle-stimulating and luteinizing hormones) in the blood flow, or ovarian regression [46].

Heat stress affected laying rate but not eggshell quality; although the hens panted, it was not enough to increase respiratory rate, decrease carbon dioxide, and cause no respiratory alkalosis or elevation of blood pH [32,47,48]. This could be corroborated because no weak or incomplete shell formation was observed.

The data from this study demonstrates that the morphological index of the egg is adequate for sales, since it is within the range of normal eggs and of good quality [24,29]. The data collected in this study, with the exception of treatment E₃₄₇₀, indicate that different levels of metabolizable energy in the diet do not affect the quality of the shell index,

which determines the hardness and permeability of the egg. Therefore, the eggs are not susceptible to microbial contamination [50]. The diet with lowest energy content shows an improved egg white index, which suggests that in warm subhumid climate conditions, the egg has better internal quality [51]. Which gives it a higher market value for its commercialization [42-45].

Egg production in high-laying hens creates enormous environmental pollution problems, mainly due to the high feed consumption that generates high production of excrement, with high levels of nitrogen and phosphorus, which can contaminate the soil and groundwater, generate greenhouse gas emissions and unpleasant odors [52].

Conclusions and Implications

High-laying Brown Nick hens, maintained in a warm subhumid climate, can produce adequately with high levels of metabolizable energy. This offers advantages because it reduces feed intake, improves some egg characteristics, and minimizes the heat generated by the birds' metabolism. Providing 3300 kcal of ME/kg of feed is a viable alternative for regulating heat stress and improving the productivity of high-laying hens. It is recommended that when using a high-energy diet, the other nutrients in the diet be adjusted (amino acids, crude protein, minerals, and vitamins). These characteristics can encourage egg production with a more sustainable use of natural resources. However, eggs produced with 2617 kcal of ME/kg have better internal quality. For future research under such conditions in which the experiment was conducted, it is suggested to include evaluations of other variables such as yolk color, Haugh units, as well as other yolk and white variables. For the raising of high-laying hens in subhumid tropics, establishing natural or artificial ventilation, installing a sprinkler system, increasing roof height, providing ample fresh water, and using additives are recommended to minimize heat stress.

Conflict of Interest Statement

We declare that we have no conflicts of interest.

References

1. Attia YA, El-Hamid Abd, Abedalla AA, Berika MA, Al-Harthi MA, et al. (2016) Laying performance, digestibility and plasma hormones in laying hens exposed to chronic heat stress as affected by betaine, vitamin C, and/or vitamin E supplementation. SpringerPlus 5: 1619.
2. SNSICA (2019) Servicio Nacional de Sanidad Inocuidad y Calidad Agroalimentaria. Manual de huevo para plato. México.

3. Castro HR (2017) Propuestas nutricionales para gallinas de postura de la línea Isa Brown, en periodo de postura, en Tingo María. Tingo, Perú: Universidad Nacional Agraria de la Selva.
4. Zarate AJ, Morán ET, Burnham DJ (2003) Exceeding essential amino acid requirements and improving their balance as a means to minimize heat stress in broilers. *J Appl Poult Res* 12(1): 37-44.
5. Felver-Gant JN, Mack LA, Dennis RL, Eicher SD, Cheng HW (2012) Genetic variations alter physiological responses following heat stress in 2 strains of laying hens. *Poult Sci* 91(7): 1542-1551.
6. Estrada MM, Márquez SM (2005) Interacción de los factores ambientales con la respuesta del comportamiento productivo en pollos de engorde. *Rev Colomb Cienc Pecu* 18(3): 246-257.
7. Balnave D, Muheereza SK (1997) Improving eggshell quality at high temperatures with dietary sodium bicarbonate. *Poult Sci* 76(4): 588-593.
8. Batres GRL (2016) Evaluación del efecto de tres niveles de energía metabolizable sobre índices productivos en aves de postura de línea genética Lohmann White. San Carlos, Guatemala; Universidad de San Carlos.
9. Rojas-Avendaño L, Arcos-González S, López-Pozos R, Santiago-Romero H, Arcos-García JL, et al. (2024) Adjustment of Amino Acids and Metabolizable Energy in Diets for High-Laying Brown Nick Hens under Heat Stress. *Int J Zoo Animal Biol* 7(6): 1-8.
10. Barzegar S, Wu S-B, Noblet J, Choct M, Swick RA (2019) Energy efficiency and net energy prediction of feed in laying hens. *Poult Sci* 98(11): 5746-5758.
11. Hwan KK, Seong BP, Jin JJ, Hyun SK, Ki TP, et al. (2018) Effect of increasing levels of apparent metabolizable energy on laying hens in barn system. *Asian-Australas J Anim Sci* 31(11): 1766-1722.
12. Harms RH, Russell BG, Sloan RD (2000) Performance of four strains of commercial layers with major changes in dietary energy. *J Appl Poult Res* 9(4): 535-541.
13. Çabuk M, Bozkurt M, Kirkpınar F, Özkul H (2001) Effect of phytase supplementation of diets with different levels of phosphorus on performance and egg quality of laying hens in hot climatic conditions. *S Afri J Anim Sci* 34(1): 13-17.
14. H&N International (2020) Brown Nick ponedoras de huevo marrón, nueva guía de Manejo. H&N International GmbH.
15. Santiago RH, Teixeira ALF, Hannas MI, Lopes DJ, Kazue SN, et al. (2017) Tablas Brasileñas para aves y cerdos, composición de alimentos y requerimientos nutricionales. Universidad Federal de Viçosa. Brazil.
16. Fan Z, Li J, Zhang Y, Wu D, Zheng X, et al. (2021) Excessive dietary lipid affecting growth performance, feed utilization, lipid deposition, and hepatopancreas lipometabolism of large-sized common carp (*Cyprinus carpio*). *Front Nutr* 13(8): 694426.
17. Google Earth (2021) Localización del proyecto en la comunidad de Bajos de Chila, Oaxaca. México.
18. INEGI (2021) Instituto Nacional de Estadística y Geografía, Climatología de México.
19. HY-Line Brown (2018) Sistemas alternativos, guía de manejo. Hy-Line International.
20. Church DC, Pond GW (1987) Eds., Fundamentos de nutrición y alimentación de animales. México: Limusa.
21. Buxadé C (2000) La gallina ponedora, sistema de explotación y técnicas de producción. España: Mundi Prensa.
22. AOAC (2000) Official methods of análisis. Association of Official Analytical Chemists. In: 17th (Edn.), Washington, DC: Association of Official Analytical Chemists.
23. Rojas VVR (2014) Uso de un aditivo a base de cantaxantina y extracto de achiote (*Bixa orellana* L.) en dietas de gallinas de postura y su efecto sobre la coloración de la yema y la vida de anaquel del huevo. Trujillo, Perú: Universidad Nacional de Trujillo.
24. De la Cruz TAF (2021) Efecto de la calidad de huevo de gallinas criollas (*Gallus domesticus*) a diferentes tiempos de conservación a temperatura ambiental en Santa Elena. La libertad, Ecuador: Universidad Estatal Península de Santa Elena.
25. Juárez-Caratachea A, Gutiérrez-Vázquez E, Segura-Correa J, Santos-Ricalde R (2010) Calidad del huevo de gallinas criollas criadas en traspatio en Michoacán, México. *Rev Cient Am Lat* 2(1): 109-115.
26. SAS (2010) SAS Education Analytical Suite for Windows (Release 9.2). Cary NC, USA: SAS Inst Inc Order number 582235. Tech support site number 70088431.
27. López KL (2018) Efecto de la formulación por aminoácidos digestibles y diferentes niveles de proteína bruta para gallinas ponderas sobre el comportamiento productivo. Trujillo, Peru: Universidad Privada Antenor Orrego.

28. Bohin E, Peh HC, Avidar Y, Israelì B, Kevkhaye E, et al. (1997) Sex and genotype dependence on effects of long-term high environmental temperatures on cellular enzyme activities from the chicken organs. *Avian Pathol* 26(3): 511- 513.
29. Mashaly MM, Hendricks GL, Kalama MA, Gehad AE, Abbas AO, et al. (2004) Effect of heat stress on production parameters and immune responses of commercial laying hens. *Poult Sci* 83(6): 889-894.
30. Cortes CA, Esparza C, Sanabria ECA, Miguel IG, Ornelas RJ, et al. (2012) El uso de granos secos de destilería con solubles (DDGS) en dietas sorgo-soya para pollos de engorda y gallinas de postura. *Rev Mex Cienc Pecu* 3(3): 331-3341.
31. Estrada PMM, Márquez GSM (2005) Interacción de los factores ambientales con la respuesta del comportamiento productivo en pollos de engorde. *Rev Col Cienc Pec* 18(3): 246-257.
32. Lara LJ, Rostagno MH (2013) Impact of Heat Stress on Poultry Production. *Animals* 3(2): 356-369.
33. Ajakaiye JJ, Perez-Bello A, Cuesta-Mazorra M, García DJR, Mollineda-Trujillo Á (2010) El estrés calórico en algunos electrolitos de plasma de gallinas ponedoras durante el verano en clima caliente-húmedo y suplementadas con vitaminas C y E. *Rev Cubana Cienc Agric* 44(4): 407-413.
34. Aberra M (2011) Performance and Physiological Responses of Naked-neck Chickens and Their F Crosses 1 with Commercial Layer Breeds to Long-Term High Ambient Temperature. *Global Vet* 6(3): 272-280.
35. Tepox PMA, Fuente MM, Benjamín JT, Ávila GE (2012) Diferentes niveles de energía metabolizable y aminoácidos azufrados en dietas para gallinas Bovans blancas. *Rev Mex Ciencias Pecu* 3(4): 439-448.
36. Vázquez GJP (2015) Efecto de cuatro tratamientos con diferente nivel de energía sobre la producción y calidad del huevo en gallinas de segundo ciclo. Aguascalientes, México: Universidad Autónoma de Aguascalientes.
37. LeClair RJ (2023) Cell Biology, genetics, and biochemistry for pre-clinical Students. Virginia Tech Carilion School of Medicine in association with Virginia Tech Publishing Blacksburg, VA.
38. Isa Brown (2009) Guía de manejo general de ponedoras comerciales. A Hendrix Genetics Company.
39. Star L, Kemp B, Van den Anker, Parmentier HK (2008) Effect of single or combined climatic and hygienic stress in four layer lines: 1. Performance. *Poult Sci* 87(6): 1022-1030.
40. Ku KH, Seong BP, Soo KH, Ho KS, Ho KC (2018) Efecto de niveles crecientes de energía metabolizable aparente en gallinas ponedoras en sistema de establo. *Asian-Australas J Anim Sci* 31(11):1766-1772.
41. Mexicana N (2016) Productos avícolas huevo fresco de gallina especificaciones y métodos de prueba. NMX-FF-127-SCFI-2016. México.
42. USDA (2000) United States Department of Agriculture, Washington. Egg-Grading manual.
43. Lacayo CAF, Miliam FBE (2013) Evaluación de las líneas de gallinas ponedoras Hy-line CV-22® y Dekalb White® en un sistema de semi-pastoreo en Zamorano, Honduras. Zamorano, Honduras: Escuela Agrícola Panamericana.
44. García CT (2015) Extracción y aplicaciones alimentarias de membranas de cáscaras de huevo. Oviedo, España: Universidad de Oviedo.
45. Vera-Rodríguez JH, Cepeda-Landin WE, Torres-Ajila KM, Bueno-Guallpa EK, Mendoza-López CA, et al. (2020) Evaluación de la calidad del huevo marrón comercial del cantón La Troncal, Ecuador. *Rev Colomb Cienc Anim* 12(2): e771.
46. Hackbart KS, Ferreira RM, Dietsche AA, Socha MT, Shaver RD, et al. (2010) Effect of dietary organic zinc, manganese, copper, and cobalt supplementation on milk production, follicular growth, embryo quality, and tissue mineral concentrations in dairy cows. *J Anim Sci* 88: 3856-3870.
47. Ajakaiye JJ, Ayo, JO, Ojo SA (2010) Effects of heat stress on some blood parameters and egg production of Shika Brown layer chickens transported by road. *Biol Res* 43: 183-189.
48. Felver-Gant JN, Mack LA, Dennis RL, Eicher SD, Cheng HW (2012) Genetic variations alter physiological responses following heat stress in 2 strains of laying hens. *Poult Sci* (91): 1542-1551.
49. Ojeda RA, Olmo GC (2022) Comparación física de la calidad externa e interna del huevo de tres razas de gallinas. *Opuntia Brava* 14(1): 134-144.
50. Juárez-Caratachea A, Gutiérrez-Vázquez E, Pérez-Sánchez RE, Román-Bravo RM, Ortiz-Rodríguez R (2011) Evaluación física de la calidad externa e interna del huevo de pavas nativas (*Melleagris gallipavo g*). *Rev Cient FCV-Luz* 21(6): 524-532.
51. Casas RS, Casas LG, Ceró A, Uña F (2016) Empleo de

los diámetros del huevo para el cálculo del volumen y superficie y su correlación con otros caracteres externos e internos en tres propósitos de gallinas reproductoras. Rev Prod Anim 28(2-3): 33-38.

52. Casas RS, Guerra CLD (2020) La gallinaza, efecto en el medio ambiente y posibilidades de reutilización. Rev Prod Anim 32(3): 87-102.