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Variation in Endogenous Gross Energy of Chickens and Its Influence on Metabolizable Energy Values of Feed Ingredients (Postprint)

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Abstract

This experiment aimed to investigate the variation in endogenous total energy of experimental chickens across different seasons and batches, and its impact on the true metabolizable energy values of feed ingredients. The experiment adopted a single-factor completely randomized design, with a total of 12 batches across three seasons (spring, summer, and autumn) to determine the endogenous total energy of chickens and its effect on the true metabolizable energy values of feed ingredients [corn, corn dried distillers grains with solubles (DDGS), dried cassava, and cassava residue], with 4 replicates per batch and 3 birds per replicate. The average value of endogenous total energy across batches within each season was used as the seasonal endogenous total energy, and its impact on the true metabolizable energy values of the four feed ingredients was calculated separately according to different seasons. The results showed: 1) The 48 h endogenous total energy differed significantly among the 12 measurement batches ($P<0.05$), but did not differ significantly among the three seasons ($P>0.05$). Therefore, the 48 h endogenous total energy data across batches within each measurement season could be combined, with the average value used as the seasonal 48 h endogenous total energy. 2) Comparing the 48 h endogenous total energy among the three seasons, the autumn 48 h endogenous total energy (67.97 kJ/bird) was significantly lower than that in spring (83.07 kJ/bird) and summer (79.90 kJ/bird) ($P<0.01$), but the difference between spring and summer was not significant ($P>0.05$). 3) The seasonal 48 h endogenous total energy was significantly positively correlated with the 48 h endogenous dry matter excretion (r$ $0.91, $P<0.01$). 4) For the four feed ingredients of corn, corn DDGS, dried cassava, and cassava residue, the maximum variation in seasonal 48 h endogenous total energy accounted for 7.36%~8.38%, 2.68%~2.94%, 7.92%~10.86%, and 3.53%~3.96% of the total energy excretion from feed, respectively, and the variation in seasonal 48 h endogenous total energy caused changes in the true metabolizable energy values of feed ingredients ranging from 0.28~0.36 kJ/g. It can be concluded that chicken endogenous total energy exhibits certain variation among seasons, but this variation does not significantly affect the calculation of true metabolizable energy values of feed ingredients.

Full Text

The Variation of Gross Energy of Endogenous Excreta and Its Effect on the Value of Metabolizable Energy of Feedstuffs in Roosters

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Abstract

This experiment was conducted to investigate the variation in gross energy of endogenous excreta (GEEE) across different seasons and batches of determination, and its effect on the true metabolizable energy (TME) values of feed ingredients in roosters. A single-factor completely randomized design was employed, with GEEE measured in 12 batches across spring, summer, and autumn seasons, using four replicates per batch and three roosters per replicate. The study examined the effects on four feed ingredients: corn, corn distillers dried grains with solubles (DDGS), dried cassava root, and cassava meal. The mean GEEE from all batches within each season was used as the seasonal GEEE value, which was then applied to calculate its impact on the TME values of the four feed ingredients.

The results showed: (1) Significant differences were observed in 48-h GEEE among the 12 determination batches ($P<0.05$), but no significant differences were found in 48-h GEEE among batches within the same season ($P>0.05$). Therefore, data from different batches within a season could be pooled, with the average value representing the seasonal 48-h GEEE. (2) Comparison of 48-h GEEE across the three seasons revealed that the autumn value (67.97 kJ/bird) was significantly lower than those in spring (83.07 kJ/bird) and summer (79.90 kJ/bird) ($P<0.01$), while no significant difference existed between spring and summer ($P>0.05$). (3) Within each season, 48-h GEEE showed an extremely significant positive correlation with 48-h endogenous dry matter excretion (r$ $0.91, $P<0.01$). (4) For the four feed ingredients, the maximum seasonal variation in 48-h GEEE accounted for 7.36%-8.38%, 2.68%-2.94%, 7.92%-10.86%, and 3.53%-3.96% of the total feed gross energy excretion for corn, corn DDGS, dried cassava root, and cassava meal, respectively. This variation in seasonal GEEE caused changes in TME values ranging from 0.28 to 0.36 kJ/g. In conclusion, roosters exhibit some seasonal variation in GEEE, but this variation does not significantly affect the calculated TME values of feed ingredients.

Key words: gross energy of endogenous excreta; true metabolizable energy; roosters

Introduction

Currently, China employs Sibbald' s [1] precision-fed cecectomized rooster assay as the national standard [2] for determining the metabolizable energy values of poultry feedstuffs. Variation in endogenous energy losses within this method may affect the reproducibility of TME determination results for feed ingredients [3]. Previous studies have indicated that variation in GEEE of experimental roosters is primarily caused by individual differences and the thermal environment of the metabolic chambers [4]. Therefore, investigating the variation in GEEE across different seasons and determination batches, and its impact on TME calculations, is crucial for evaluating the stability of the national standard method for determining TME values of feed ingredients in chickens.

Sibbald et al. [5-6] summarized data from 38 metabolic trials and reported that the 48-h GEEE of adult White Leghorn roosters ranged from 49.96 to 188.62 kJ/bird. Seven European laboratories found that the 48-h GEEE of adult Rhode Island Red roosters ranged from 18.8 to 87.8 kJ/bird [7]. He and Xiao [8] reported that the 48-h GEEE of 9-week-old Avian roosters ranged from 98.73 to 351.08 kJ/bird. These findings demonstrate substantial variation in GEEE even within the same breed. In the precision-fed assay, the ratio of GEEE variation to total feed intake directly determines the magnitude of variation in calculated TME values [9]. If this ratio is low (<0.42 kJ/g), the impact of GEEE variation on calculated TME values of test feeds is considered minor; conversely, GEEE variation must be carefully considered in TME determination. Accordingly, this study used adult Hy-Line Brown roosters to measure the variation in GEEE across different seasons and batches, and its effect on calculated TME values of four feed ingredients, to investigate whether GEEE variation in the current national standard (GB/T 20437-2010) for poultry feed energy evaluation affects the reproducibility of TME results and to provide references for further standardizing the application of this method.

Materials and Methods

1.1 Experimental Design and Animals

This study consisted of two experiments. Experiment 1 investigated the variation in body weight changes and GEEE of roosters across different seasons and batches using the national standard method (GB/T 26437—2010). A single-factor completely randomized design was employed with 108 adult Hy-Line Brown roosters of uniform body weight [(2.12$±$0.32) kg] and normal behavior, randomly divided into 9 groups with 4 replicates per group and 3 roosters per replicate. For each batch of GEEE determination, one group was randomly selected: in spring, groups 9, 2, 5, 1, 8, and 3 were selected sequentially; in summer, groups 6, 4, 1, and 9 were used; and in autumn, groups 2 and 7 were selected to measure 48-h endogenous dry matter excretion, 48-h GEEE, and body weight changes during the test period.

Experiment 2 examined the effect of GEEE variation on TME values of feed ingredients. According to the national standard for poultry feed energy determination (GB/T 20437—2010), the apparent metabolizable energy values of three corn samples, three corn DDGS samples, three dried cassava root samples, and three cassava meal samples were determined, with 4 replicates per feed sample and 3 roosters per replicate. Based on the seasonal GEEE values calculated from Experiment 1 (spring, summer, and autumn), the TME values of each feed ingredient were calculated using the respective seasonal GEEE corrections to assess the impact of GEEE variation on TME values. During metabolic trials, roosters were managed according to the standard operating procedures of GB/T 26437—2010, with metabolic chamber temperatures maintained at 10-27 °C, natural lighting, natural ventilation, and free access to water.

The four test feed ingredients were provided by New Hope Liuhe Feed Co., Ltd., with their nutrient contents shown in Table 1 .

1.3 Metabolizable Energy Determination Method

The metabolizable energy determination procedure followed the national standard "Technical Specification for Determination of Apparent Metabolizable Energy of Chicken Feed by Precision Feeding Method" (GB/T 26437—2010). The protocol consisted of a 3-day pre-test period during which roosters were fed a corn-soybean meal basal diet, with the final meal consisting of the test feed ingredient (the endogenous group continued receiving the basal diet). Following the pre-test period, feed was withheld for 48 h. Roosters were then precision-fed 50 g of test feed ingredient (weighed to 0.0002 g precision), while the endogenous group remained fasted, with all birds having free access to water.

Excreta collection and processing: Excreta collection followed the methods specified in GB/T 26437—2010. When collection bags exceeded one-third capacity, excreta were transferred without loss to corresponding numbered glass petri dishes. After collection, dishes were immediately placed in a 65 °C oven for drying, followed by 24 h equilibration at room temperature before weighing and recording dry matter content (determined according to GB/T 6435—2006). Upon completion of the metabolic trial, gross energy of excreta was determined following ISO 9831:1998 procedures, with dry matter content measured simultaneously. Metabolizable energy was calculated using the following formulas:

$$\text{Apparent Metabolizable Energy (AME)} = \frac{\text{Feed DM intake} \times \text{Feed GE} - \text{Excreta DM} \times \text{Excreta GE}}{\text{Feed DM intake}}$$

$$\text{True Metabolizable Energy (TME)} = \frac{\text{Feed DM intake} \times \text{Feed GE} - \text{Excreta DM} \times \text{Excreta GE} + \text{Endogenou}}{\text{Feed DM intake}}$$

1.4 Data Processing and Statistical Analysis

The PROC GLM module in SAS 9.3 was used for analysis of variance on body weight changes, endogenous dry matter excretion, GEEE, and other parameters across all determination batches and between spring and summer fasting conditions, with Duncan’ s multiple range test for post-hoc comparisons. Data from the two autumn batches were compared using t-tests. The CONTRAST module was used for variance analysis of body weight changes, endogenous dry matter excretion, and GEEE across seasons under fasting conditions. The PROC CORR module was used to analyze correlations between GEEE and body weight changes and endogenous dry matter excretion across different batches within seasons. Significance was declared at $P<0.05$ and extreme significance at $P<0.01$.

Results

2.1 Variation in Gross Energy of Endogenous Excreta Across Batches

As shown in Table 2 , significant differences were observed among the 12 determination batches in pre-test body weight, post-test body weight, body weight loss, 48-h endogenous dry matter excretion, energy content per gram of endogenous dry matter, and 48-h GEEE ($P<0.05$). Within the six spring batches, significant differences were found in pre-test body weight, post-test body weight, body weight loss, and energy per gram of endogenous dry matter ($P<0.05$), but not in 48-h endogenous dry matter excretion or 48-h GEEE ($P>0.05$). In the four summer batches, no significant differences were observed in pre-test body weight, post-test body weight, body weight loss, or 48-h GEEE ($P>0.05$), while 48-h endogenous dry matter excretion and energy per gram of endogenous dry matter differed significantly ($P<0.05$). In the two autumn batches, no significant differences were detected in any measured parameters ($P>0.05$).

Between seasons, spring and summer showed no significant differences in pre-test body weight, post-test body weight, or 48-h GEEE ($P>0.05$), but exhibited significant differences in body weight loss, 48-h endogenous dry matter excretion, and energy per gram of endogenous dry matter ($P<0.05$). Spring and autumn differed significantly in pre-test body weight, post-test body weight, body weight loss, energy per gram of endogenous dry matter, and 48-h GEEE ($P<0.05$), but not in 48-h endogenous dry matter excretion ($P>0.05$). Summer and autumn showed no significant differences in body weight loss or 48-h endogenous dry matter excretion (P>0.05), but differed extremely significantly in pre-test body weight, post-test body weight, energy per gram of endogenous dry matter, and 48-h GEEE (P<0.01).

2.2 Correlation Between Gross Energy of Endogenous Excreta and Body Weight

As presented in Table 3 , across spring, summer, and autumn seasons, 48-h GEEE showed no significant correlation with pre-test body weight, post-test body weight, body weight loss, or energy per gram of endogenous dry matter (P>0.05), but demonstrated a highly significant positive correlation with 48-h endogenous dry matter excretion (r\$ $0.91, P < 0.01). Whenanalyzingalldatafromthe12batchescollectively, although48 - hGEEEshowedsignificantcorrelationswithpre - testbodyweight, post - testbodyweight, andenergypergramofendogenousdrymatter(P < 0.05), thecorrelationcoefficientswerelow(r hendogenousdrymatterexcretionremainedhigh(r$ \$0.71, P<0.01).

2.3 Effect of Gross Energy of Endogenous Excreta on Metabolizable Energy Values of Feed Ingredients

As shown in Table 2, no significant differences were observed in 48-h GEEE among batches within each season (P>0.05), though significant differences existed between seasons (P<0.05). Therefore, 48-h GEEE data from different batches within the same season were pooled, and the average values were used as the seasonal 48-h GEEE.

Table 4 presents the effects on feed ingredients. For the three corn samples, 48-h feed gross energy excretion ranged from 139.92 to 178.05 kJ/bird, with endogenous energy accounting for 39.34%-59.45% of feed gross energy excretion. The maximum seasonal variation in GEEE represented 7.36%-8.38% of feed gross energy excretion, causing TME value variations of 0.28-0.29 kJ/g. For the three corn DDGS samples, 48-h feed gross energy excretion ranged from 446.16 to 488.29 kJ/bird, with endogenous energy comprising 14.34%-18.63% of feed gross energy excretion. Maximum seasonal GEEE variation accounted for 2.68%-2.94% of feed gross energy excretion, resulting in TME value variations of 0.29-0.30 kJ/g.

For the three dried cassava root samples, 48-h feed gross energy excretion ranged from 123.53 to 165.29 kJ/bird, with endogenous energy representing 42.35%-67.60% of feed gross energy excretion. Maximum seasonal GEEE variation constituted 7.92%-10.86% of feed gross energy excretion, causing TME value variations of 0.29 kJ/g. For the three cassava meal samples, 48-h feed gross energy excretion ranged from 331.27 to 371.03 kJ/bird, with endogenous energy accounting for 18.87%-25.08% of feed gross energy excretion. Maximum seasonal GEEE variation represented 3.53%-3.96% of feed gross energy excretion, resulting in TME value variations of 0.29-0.36 kJ/g.

Discussion

3.1 Factors Influencing Variation in Gross Energy of Endogenous Excreta

In Sibbald' s precision-fed cecectomized rooster assay, endogenous excreta primarily originate from secreted bile, digestive fluids, and sloughed intestinal epithelial cells, representing part of the animal' s maintenance metabolism [5,10]. Early studies demonstrated substantial individual variation in GEEE within the same flock, with coefficients of variation ranging from 1.95% to 15.88% [11]. More recently, Fan et al. [12] reported a mean 48-h GEEE of 88.88 kJ/bird with a coefficient of variation of 9.55% in 48 Hy-Line Brown roosters. Ren [13] found 48-h GEEE values of 70.13–77.24 kJ/bird with coefficients of variation of 4.11%-10.77% in yellow-feathered broilers. In the present study, 48-h GEEE in Hy-Line Brown roosters across 12 batches ranged from 67.01 to 91.20 kJ/bird, with coefficients of variation of 3.15%–15.32%. These data further confirm that substantial individual variation in GEEE within the same breed is an objective reality.

Among factors influencing GEEE, body weight of the same breed does not show a stable correlation with GEEE [14]. Our results also indicated that 48-h GEEE was not significantly correlated with pre-test body weight, post-test body weight, body weight loss, or energy per gram of endogenous dry matter during metabolic trials, consistent with our laboratory's previous findings in yellow-feathered broilers. However, environmental temperature during metabolic trials significantly affects GEEE, with GEEE decreasing as temperature increases [15-16]. In this study, metabolic chambers were heated to maintain 10–15 °C in winter and spring, air-conditioned to 20–25 °C in summer, but no temperature control was implemented in autumn. Based on Beijing' s climate characteristics, autumn chamber temperatures (September–October) were higher than in spring and summer, resulting in lower GEEE values in autumn. This suggests that artificial environmental control should be continued in autumn under our laboratory conditions to reduce GEEE fluctuations.

3.2 Effect of GEEE Variation on Metabolizable Energy Values of Feed Ingredients

In the precision-fed assay, both individual roosters and the same rooster across different trials exhibit substantial variation in GEEE [6]. Although this variation coefficient can be as high as 10%, its impact on the TME value of test feed ingredients does not exceed 1% [10], while the maximum difference in feed ingredient TME values between batches is approximately 0.42 kJ/g, equivalent to 3% of the metabolizable energy value [17]. This indicates that despite large variation in GEEE itself, the total change in feed TME values caused by GEEE variation remains within 0.42 kJ/g. Therefore, feed TME values are primarily related to feed intake or the inherent energy value of the feed itself.

In this study, no significant differences in 48-h GEEE were observed among batches within seasons (maximum difference of 11.99 kJ/bird), which translated to a maximum change in TME values of approximately 0.27 kJ/g per gram of air-dry feed. Although significant differences existed between spring/summer and autumn GEEE (maximum difference of 13.10 kJ/bird), this translated to a maximum TME value change of approximately 0.36 kJ/g per gram of air-dry feed. This indicates that while significant seasonal differences in GEEE exist, their impact on TME values does not exceed the precision of the precision-fed assay (0.42 kJ/g). Therefore, the effect of GEEE variation on TME values is limited.

However, the proportion of GEEE in total feed gross energy excretion varies among feed ingredients. In this study, GEEE accounted for over 39.34% and 42.35% of feed gross energy excretion in corn and dried cassava root, respectively, but less than 18.63% and 25.08% in corn DDGS and cassava meal, respectively. This suggests that GEEE variation contributes more substantially to variation in feed gross energy excretion for corn and dried cassava root than for corn DDGS and cassava meal, potentially influencing the variation in TME values accordingly.

Conclusion

Based on the findings of this study, three main conclusions can be drawn. First, no significant differences in GEEE were observed among different determination batches within spring, summer, and autumn seasons, though GEEE values in spring and summer were significantly higher than in autumn. Second, the maximum difference in 48-h GEEE between batches within seasons and across seasons was within 13.10 kJ/bird, causing changes in TME values of corn, corn DDGS, dried cassava root, and cassava meal within 0.36 kJ/g. Third, although seasonal variation in GEEE exists in roosters, this variation does not significantly affect the calculated TME values of feed ingredients, remaining within the acceptable precision range of the precision-fed assay method.

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