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## Use of LIPE® as External Indicator, Replacing the Total Collection for Measuring the Coefficients of Metabolization of Nutrients and Metabolizable Energy for Broilers

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**Abstract:** To evaluate the efficacy of the LIPE® as external marker to measure the digestibility of nutrients and Apparent Metabolizable Energy (AME) and apparent metabolizable corrected for nitrogen balance (AMEn) compared to the total collection method for broilers from one to 21 days and 22 to 42 days, 15 chicks one day old and 10 broilers with 22 days of age, respectively, in a randomized block design with two treatments and 24 repetitions, each repetition being considered as a block. There was no significant correlation ( $P>0.05$ ) for the metabolizable coefficient of dry matter (MDM) for both phases. For the metabolizable coefficient of crude protein (MCP) in the initial phase, the correlation was median (between 30 and 70%), positive and significant ( $P\leq 0.01$ ), while for the metabolizable coefficient of ether extract (MEE) and AME and AMEn, the correlations were strong (above 70%), positive and significant ( $P\leq 0.01$ ), as well as to the MCP, MEE, AME and AMEn in the growth phase in which strong correlations were found (above 70%), positive and significant ( $P\leq 0.01$ ). When comparing the same variables using analysis of variance, depending on the methodology employed, there was no difference by F test ( $P>0.05$ ) for MDM, MCP, MEE, AME and AMEn for both phases, with the exception of AMEn in the initial phase, in which a higher value was observed ( $P\leq 0.05$ ) for LIPE®. It is concluded that the LIPE® as external indicator can be used to measure the production of excreta and subsequent determination of metabolization coefficient and energy values for broilers.

**Key words:** Metabolization of nutrients, broiler, indicator, LIPE®

### INTRODUCTION

The conventional method of total excreta collection was proposed to evaluate the apparent metabolization of nutrients and nutritional value of foods, providing reliable results for the intended purposes, however, according to some authors, it is a time consuming and expensive method. Because of this, new studies are being conducted to develop and use indicators that have reliable results. According to McNab (2000) during the total collection problems may occur such as fecal material adherence to the feathers of birds, contamination of excreta with feathers and dander, change in composition due to fermentation of excreta, excretion out of trays and feed contamination by regurgitated, factors that may affect the determination of nutrient digestibility.

Saliba *et al.* (1999) revealed that the lignin obtained by the extraction process had physicochemical properties quite stable and high chemical and structural consistency, showing unchanged through the gastrointestinal tract of animals and fully recovered in faeces. Research related to the study of structural and chemical composition of the physicochemical properties

of lignin have been undertaken in the best interests of its elimination as a contaminant of pulp, for the paper industry (Vasconcellos, 2004).

Thus arose the possibility of using purified lignin eucalyptus (LIPE®) as an external indicator to assess the digestibility of food by measuring the concentration of LIPE® in feed and excreta which allows us to estimate the values of consumption food and even the production of excreta.

To assess the reliability of an indicator is required to be made metabolization trials, comparing the results with those obtained by the indicator method of total excreta collection. Thus, the objective of this study was to evaluate the efficacy of the use of LIPE® as external marker to measure the metabolizable coefficient of nutrients and energy values when compared to the total collection method for broilers from one to 21 (starter phase) and 22 to 42 days of age (growth phase).

### MATERIALS AND METHODS

The metabolization of nutrients using LIPE® was measured in Laboratório de Metabolismo Animal da Escola de Veterinária da Universidade Federal de Minas

Gerais, Brazil, with broilers from 15 to 18 and 30 to 33 days of age, housed 15 Ross broilers, males, one days of age with a mean weight of 39.1±0.8 g in each of the 30 experimental units, totaling 450 birds for the starter phase and 10 Ross broilers, males, with 22 days of age, weighing average 1.038±0.030 kg in each of the 30 experimental units, totaling 300 birds for the growth phase. All procedures adopted throughout this study avoided unnecessary animal discomfort and were managed according to the directives of the Ethics and Research Committee of the Universidade Federal de Minas Gerais, Belo Horizonte, Brazil.

The incorporation of LIPE® was made by dilution in acetone and sprayed on the ration of each treatment, homogenized and letting the acetone evaporate for a period of 24 hours prior to delivery of feed to animals. The concentration of LIPE® used was 0.1 g of LIPE® for every 1 kg of feed. Were offered 0.5 kg and 1.0 kg of feed per day for a period of five days and for the starter and growth phase, respectively and two days of adaptation and three days of collection. The ration was treated with LIPE® supplied in the morning after the withdrawal of the previous day's leftovers. By late morning the remains were heavy and offered ad libitum diet without indicator. The quantities of feed offered and leftovers were weighed at the beginning and end of the experiment, respectively and the excreta collected twice a day.

The material collected was packed in plastic bags, weighed and stored in a freezer until the final period of data collection. Later the excreta were weighed and placed in an oven of forced ventilation at 60°C for 72 hours for pre-drying and soon after exposed for two hours at room temperature and then weighed and homogenized for a sample for determination of dry matter, ether extract and nitrogen. The experimental diets were also subjected to the analysis, according to techniques described by Compêndio Brasileiro de Nutrição Animal (2005).

The technique used to analyze the concentration of LIPE® in the stools and excreta was the Infrared Spectroscopy Fourier Transform, described by Rodrigues *et al.* (2007) and Excreta Production (EP) determined through the use of an external indicator by the formula:

$$EP \left( \frac{g}{dia} \text{ of DM} \right) = \frac{A.I. \text{ administered } \left( \frac{g}{dia} \right)}{A.C. \text{ of F.I. } \left( \frac{g}{deDM} \right)} \times RR$$

where:

A.I. is the Amount of Indicator

A.C. is the Average Concentration

F.I. is the Faecal Indicator

RR is the Recovery Rate estimated by the indicator

$$RR = \frac{\text{Total amount of the faecal indicator (g)}}{\text{Total amount of the indicator administered (g)}}$$

From the results of laboratory tests, together with the data of feed intake and excreta production predicted by the analysis of LIPE® as well as the total collection, we calculated the metabolizable coefficients of dry matter (MDM), crude protein (MCP) and ether extract (MEE), according to the formula below:

$$MN (\%) = \frac{\text{Nutrient intake(g)} - \text{Nutrients from excretas (g)}}{\text{Nutrient intake (g)}} \times 100$$

Where: M.N. = Metabolizable of nutrients

To compare the two methods of determining the metabolization of nutrients (total collection or use of LIPE®), the experimental design was randomized blocks with 24 treatments and two replicates of 15 chickens for the starter phase and 10 broilers for growth phase, with repetition being considered blocks. It was made the Pearson correlation between the responses of the two methods, statistical analysis made by SAS (2001).

## RESULTS AND DISCUSSION

The Table 1 shows the correlation values between the methods of determining the metabolization (LIPE® and total collection) to the responses studied.

For the variable CDMS was not observed correlation between the methodologies, for both phases, even with values for the MDM was very close numerically. For the starter phase, the variable MCP a median correlation was found (between 30 and 70%), positive and significant ( $P \leq 0.01$ ). For the other variables (MEE, EMA and AMEn), the correlations were strong (above 70%), positive and also significant ( $P \leq 0.01$ ). As in the starter phase, the correlations found for MCP, MEE, EMA and EMAN were strong (above 70%), positive and significant ( $P \leq 0.01$ ), showing the similarity and accuracy between the two methods in the measurement of coefficients metabolization and energy values for broilers.

When comparing the same variables by analysis of variance, we can observe that there were no significant differences ( $P > 0.05$ ) between the methods for the variables studied, except AMEn for starter phase as can be seen in Table 2 as well as the growth phase which can be observed that there was also no significant differences ( $P > 0.05$ ) between methods for MDM, MCP, MEE, AME and AMEn, as shown in Table 3.

In Table 2, the use of LIPE® to determine the metabolization coefficient of nutrients and energy values for broilers in the starter phase showed similar results to the method of total excreta collection, except for the value of AMEn. This difference found between the two methodologies can be coming from the measured value of nitrogen due to high variability of the MCP that may have had a cumulative effect on AMEn since the amount of nitrogen is taken into account in the equation.

In Table 3, the use of LIPE® to determine the metabolizable coefficient of nutrients and energy values

Table 1: Pearson Correlation (%) between the values of the metabolizable coefficient of dry matter (MDM), of crude protein (MCP), of ether extract (MEE), apparent metabolizable energy (AME) and apparent metabolizable energy corrected for nitrogen balance (AMEn) determined by total collection method and the use of purified lignin eucalyptus (LIPE®) for broilers

Total collection	Purified lignin eucalyptus (LIPE®)				
	MDM	MPB	MEE	EMA	EMAn
Starter phase (1 to 21 days old)	-	67.35	90.06	99.45	99.34
Growth phase (22 to 42 days old)	-	88.02	93.53	93.67	94.91

Correlation values presented are significant at  $P \leq 0.01$

Table 2: Metabolizable coefficient ( $\pm$  standard deviation) of dry matter (MDM), metabolizable coefficient of crude protein (MCP), metabolizable coefficient of ether extract (MEE), apparent metabolizable energy (AME) and apparent metabolizable energy corrected for nitrogen balance (AMEn) according to the methodologies used for the starter phase

Methodologies	MDM (%)	MCP (%)	MEE (%)	EMA (Kcal/Kg)	EMAn (Kcal/Kg)
Total collection	74.09 $\pm$ 1.69a	55.03 $\pm$ 4.69a	92.96 $\pm$ 1.45a	3,679.74 $\pm$ 103.33a	3,446.58 $\pm$ 96.32b
LIPE®	73.88 $\pm$ 1.39a	54.57 $\pm$ 3.67a	92.83 $\pm$ 1.34a	3,707.87 $\pm$ 59.09a	3,562.24 $\pm$ 63.98a
CV (%)	2.19	8.64	1.55	3.66	3.68

Means followed by different letters in the column differ by the F test ( $P \leq 0.05$ )

Table 3: Metabolizable coefficient ( $\pm$  standard deviation) of dry matter (MDM), metabolizable coefficient of crude protein (MCP), metabolizable coefficient of ether extract (MEE), apparent metabolizable energy (AME) and apparent metabolizable energy corrected for nitrogen balance (AMEn) according to the methodologies used for the growth phase

Methodologies	MDM (%)	MCP (%)	MEE (%)	EMA (Kcal/Kg)	EMAn (Kcal/Kg)
Total collection	75.33 $\pm$ 1.16	57.47 $\pm$ 4.01	94.19 $\pm$ 1.15	3,763.06 $\pm$ 136.88	3,624.07 $\pm$ 135.25
LIPE®	75.77 $\pm$ 1.26	58.09 $\pm$ 4.35	94.20 $\pm$ 1.09	3,777.94 $\pm$ 119.55	3,637.36 $\pm$ 119.18
CV (%)	1.61	7.45	1.16	3.81	3.92

Means followed by different letters in the column differ by the F test ( $P \leq 0.05$ )

for broilers during growth phase showed similar results to the method of total excreta collection, results similar to those found by Vasconcellos *et al.* (2007).

Cortés *et al.* (2009) found contrary to the results presented in this paper. They found that the use of  $\text{Cr}_2\text{O}_3$  as an indicator, underestimates the values of some coefficients metabolization compared to the total collection method, resulting in a lower recovery rate.

Despite the evidence available in the literature regarding the efficiency of use of the indicator in determining the metabolization of nutrients, are not fully conclusive, some authors (Yoshba and Morimoto, 1957; Han *et al.*, 1976; Vasconcellos *et al.*, 2007) found that the use of external indicators show to be effective and equivalent to the total collection, as well as the present work.

**Conclusion:** The use of LIPE® as external indicator, in circumstances in which this experiment was done, for the measurement of fecal production and subsequent determination of the coefficients of metabolization and energy values for broilers is efficient and equivalent to the total collection.

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