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Evaluation of the Metabolizable Energy of Meat and Bone Meal for Chickens and Turkeys by Various Methods

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Abstract: A series of assays were conducted to determine to the available energy of several meat and bone meal products by various methods. The correct determination of the energy available to poultry from these products is important for accurate diet formulation and product usage. Several methods for determining energy availability are utilized, including true metabolizable energy (TME) and excreta and digesta apparent metabolizable energy (AME). The goal of these experiments was to determine if any differences exist among the available energy values determined by the various methods. There were few differences in assay methodologies noted. There were significant differences in mean ME values among products, as would be expected due to meat and bone meal's variable nature. Chick digesta AME values were usually the only cause of difference, typically lower than other values. There were no differences between pooled AME and TME values, or between chickens and turkeys. In general, it appears that the TME values commonly determined with Leghorn roosters are acceptable for broilers and turkeys. The proximate analysis and mineral composition values of the meat and bone meal products were also determined. These values were then used to develop equations that predict the TME_n of a product. The products were too variable to provide an accurate equation using the proximate values (R^2 = 0.42). Using the gross energy of the product as a predictor variable greatly improved the accuracy, with an R^2 of 0.77.

Key words: Metabolizable energy, meat and bone meal, prediction equation, turkeys

Introduction

Meat and bone meal is a common by-product used in poultry rations. It is typically composed of rendered beef or pork materials, containing approximately 50% crude protein, 8% fat, 28% ash, 10% calcium, and 5% phosphorus. With recent bans on the feeding of ruminant tissues to ruminants, meat and bone meal may find increased usage in poultry rations. Unfortunately, the quality of meals varies greatly, making it difficult to precisely measure the nutrient availability (Elkin, 2002). Proper utilization of meat and bone meal requires accurate metabolizable energy (ME) values that can be determined in a timely manner. In order to maximize utilization of meat and bone meal products, the nutritive value of these feedstuffs must be readily assessed. While protein, moisture, fat, and others components can be quickly ascertained via proximate analysis, metabolizable energy (ME) requires more elaborate assays. Determining ME requires the use of test animals, sample collection, and bomb calorimetry. This process can take several weeks.

Most energy values are based on Leghorn rooster evaluations, though species differences have been shown (Farhat *et al*., 1998; Ostrowski-Meissner, 1984). The first objective of this study was to compare the energy availability of several meat and bone meal products for both chickens and turkeys using several different energy measurement systems. The second objective was to develop an equation that nutritionists could use to rapidly determine the nutritive value of a meat and bone meal product.

Materials and Methods

A total of 12 meat and bone meals were obtained through commercial sources. The proximate composition and mineral composition of each sample was determined (AOAC, 1970). Each product was subjected to a series of assays similar to those of Zanella *et al*. (1999). Zanella *et al*. (1999) studied enzyme enhanced digestibility. There were four assays conducted with both chickens and turkeys for a total of eight treatments, a 2x4 factorial design. The first assay was based on Sibbald's TME system (1986). Modifications were made for cecectomized roosters and intact turkeys. For both species, birds were not allowed feed for 36 hours, instead of 24 hours, to ensure adequate clearing of the gastrointestinal tract. They were then tube fed a measured quantity of product, 30 grams for roosters and 75 grams for turkeys and placed in metabolism cages. Each product was replicated eight times as were birds used for endogenous collection. Excreta were then collected for 48 hours. Excreta were then dried at 60°C in a forced air oven and weighed. The gross energy of the feed and the excreta content were determined via bomb calorimetry. Nitrogen content of feed and excreta were also determined by LECO analysis (AOAC Method 990.03) for nitrogen correction. An example of the calculation for TME follows:

$$TME = GE_{feed} - GE_{fed\ excreta} + GE_{fasted\ excreta}$$

Where the GE of the excreta from fasted birds was used to correct for endogenous loss. Excreta were nitrogen corrected to maintain a zero nitrogen balance offset from fasting and yield a TME_n value.

The second two assays were designed based on the ME system to determine apparent metabolizable energy (AME) and AME adjusted for endogenous loss (aAME) via battery studies. Four-hundred and twenty commercial strain broilers and 420 commercial turkey hens were obtained at the day of hatch and reared to 24 days of age. A basal diet was calculated (Table 1) to meet all nutritional needs for both chicks and poults recommended by the National Research Council (1994). Chicks and poults were raised in multi-tiered wire floor batteries. They were allowed feed and water *ad libitum* with constant lighting. Each pen held five birds. Feed was removed 24 hours prior to the start of the assay to allow for clearing of the gastrointestinal tract. The basal diet was then diluted with the addition of each product at 50%. On day 21, birds were allocated to pens at random and treatments were assigned via a random number table to begin the experiment. There were six replicate pens per treatment plus six pens receiving the undiluted basal diet. Another six pens were withheld from feed with total endogenous excreta collected to adjust for endogenous loss. The total feed intake and total excreta were measured for all pens. Feed and excreta were also nitrogen corrected for uniformity. The experiment was concluded after three days, at 24 days of age. The AME and aAME were calculated as follows:

$$AME = GE_{feed} - GE_{fed\ excreta}$$

$$aAME = GE_{feed} - GE_{fed\ excreta} + GE_{fasted\ excreta}$$

The energy determinations of feed and excreta were adjusted for the basal diet energy contribution in a manner suggested by Sibbald and Slinger (1963). Briefly, the ME of the basal diet accounted for half of the ME of each treatment. The ME of each product was then calculated.

The last assay was based on ileal collection in determining AME. At the end of the trial, all birds were euthanized by cardiac puncture with a sodium phenobarbital solution to prevent movement of digesta in the gut. A sodium phenobarbital solution depresses the central nervous system and therefore intestinal contractions (Barnhart, 1990). The ileal contents were collected from Meckel's diverticulum to the ileocolic juncture. Meckel's diverticulum was chosen because it is considered the end of the jejunum and the start of the ileum. Most digestion and absorption of carbohydrates, proteins, and fats occur in the duodenum and jejunum. Microbial fermentation occurs after exiting the ileum. Therefore, ileal contents should provide an adequate measure of metabolizability of a product (Scanes *et al.*, 2004). Chromic oxide was added at 0.05% of the diet as a marker. All samples were pooled by pens. Feed and digesta were also nitrogen corrected for uniformity. The following equation was used to determine digesta AME:

$$AME = GE_{diet} - [GE_{digesta} \times (Marker_{diet}/Marker_{digesta})]$$

Analysis of variance was conducted on assay methods for each product, pooled AME and TME values, pooled chicken and turkey values, and pooled digesta and excreta values. A Tukey-Kramer test was used to determine differences in means when appropriate. The level of significance was set at 0.05.

Once all data were compiled (Table 2 and 3), a multiple regression equation was used for prediction of the TME_n value of a meat and bone meal given the nutrient composition. Crude protein (CP), moisture, ash, fat, carbohydrates (CHO), gross energy (GE), calcium (Ca), phosphorus (P), sodium (Na), potassium (K), and iron (Fe) were used as predictor variables. Stepwise regression was used to determine which predictor variables were significant for a prediction equation. All data were analyzed with the JMP version of SAS.

All procedures complied with the laboratory's Standard Operating Procedures and the University of Missouri's Animal Care and Use guidelines.

Table 1: Composition of Basal Diet[1] for Chicks and Poults

Ingredients	Basal Diet %
Ground Corn	74.105
Soybean Meal (48%)	21.790
Dicalcium Phosphate	1.813
Limestone	1.485
Salt	0.250
DL-Methionine	0.009
Trace Mineral Premix[2]	0.100
Vitamin Premix[3]	0.075
Selenium Premix[4]	0.030
Choline Chloride	0.182
Copper Sulfate	0.013
Coban	0.075
Chromic Oxide	0.100

[1]Diluted to 50% with addition of MBM sample for ME assays.
[2]Trace mineral premix analysis: Ca 2.50%, Fe 6.0%, Mg 2.68%, Mn 11.0%, Zn 11.0%, I 2,000 ppm.
[3]Vitamin premix provided per kilogram of diet: Vitamin A 1,500 IU, D 200 IU, E 10 IU, K 2 mg, Thiamin 1.8 mg, Riboflavin 4.5 mg, Pyridoxine 3.5 mg, Folic acid 0.55 mg, Niacin 35 mg, Pantothenic acid 14 mg, Choline 1,300 mg.
[4]Selenium premix analysis: Ca 36.08%, Se 0.06%.

Results and Discussion

There were significant differences in the ME values obtained by different measurements in six of the 12 meat and bone meal products. Chick digesta values were typically lower than other measures when differences were noted. Turkey digesta values appear to largely agree with other measurements (Table 3). There were no differences between true and apparent metabolizable energy measurements (Table 4). The

Table 2: Proximate and Mineral Composition of Meat and Bone Meal Products

Sample	CP %	Moisture %	Ash %	Fat %	Ca %	CHO %	P %	K %	Na %	Fe ppm	GE Kcal/kg
Mbm-2	53.82	6.21	27.27	10.80	8.33	1.90	3.97	0.42	0.83	1,265	3,880
Mbm-3	50.44	8.16	28.89	10.27	9.29	2.24	4.74	0.46	1.05	381	4,130
Mbm-5	50.88	6.51	31.06	9.65	10.26	1.90	4.53	0.44	0.85	365	4,200
Mbm-7	58.97	7.94	23.85	8.37	8.08	0.87	4.32	0.54	0.68	258	4,439
Mbm-8	58.50	4.42	23.33	12.26	8.26	1.49	4.17	0.63	0.71	437	4,147
Mbm-9	51.94	5.75	27.01	12.44	9.04	2.86	4.51	0.59	1.14	456	4,347
mbm-10	51.10	3.50	26.80	11.50	*	*	*	*	*	*	3,239
mbm-12	48.32	6.32	27.32	8.17	11.82	9.87	5.90	0.40	0.92	564	3,516
mbm-13	52.44	7.83	23.62	12.12	7.45	3.99	3.80	0.64	1.04	724	3,728
mbm-14	50.07	5.03	34.20	8.57	12.09	2.13	6.03	0.47	1.11	228	3,779
mbm-15	58.94	5.17	25.71	9.41	8.93	0.77	4.60	0.63	0.88	401	4,349
mbm-16	45.69	7.04	37.73	9.00	13.59	0.54	6.83	0.33	1.10	226	3,077

*Data unavailable.

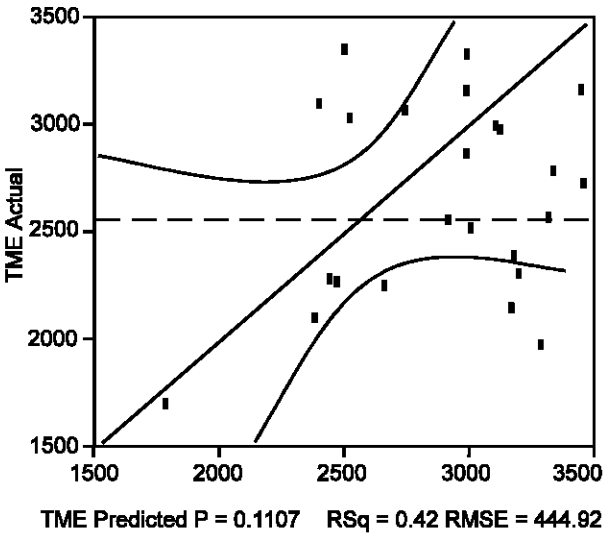

Fig. 1: Best-Fit Prediction Equation of TME_n Value of Meat and Bone Meal from Proximate Analysis.
$TME_n = 240.8 - 75.9*(CHO) + 47.8*(CP)$

pooled digesta value (2,409 kcal/kg) was significantly lower than pooled excreta value of 2,560 kcal/kg (Table 4). This was evident in some individual products as well. There were also no differences in the values obtained from chickens and those from turkeys (Table 4).

Overall, the ME values of meat and bone meal appear slightly higher than a TME_n of 2,495 kcal/kg suggested by the National Research Council (1994). The mean value of all measurements of all meat and bone meal products was 2,522 kcal/kg. There were few differences in methodology for determining metabolizable energy. There was a significant difference among assay methods for six of the 12 products. Lower chick digesta AME values accounted for most, but not all of the differences seen among methods (Table 3). Other values were relatively consistent. This could be expected given other researchers have found the use of markers can lead to inaccurate results, especially when using chromic oxide (Schneider and Flatt, 1975; National Research Council, 1994; Scott and Boldaji, 1997; Scott and Hall, 1998). Still, there are problems with the total collection of excreta that use of a marker alleviates (Sales and Janssens, 2003). However, there were still no differences among methods for six of the twelve products, indicating that the use of a marker with digesta collection may be inconsistent. There were problems obtaining enough digesta sample from some pens to complete all of the necessary procedures. These pens were not included in the statistical analysis. Having fewer replications may have aided in finding differences among methods.

There appears to be no difference between the battery ME System of Anderson *et al.* (1958) and Sibbald's TME System (1986) (Table 4). This would indicate that tube feeding may be used in place of battery trials and still obtain similar results. Dale and Fuller (1982) found that TME values are an adequate measure of metabolizable energy values. The TME System has the advantage of being less expensive to conduct, using less feed, fewer animals, and taking much less time.

Interestingly, the pooled ME values of digesta were significantly lower than excreta samples from the same experiment (Table 4), as seen with several individual products. This may be due to the reasons mentioned previously, there have been mixed reviews on the reliability of markers for metabolism assays. Also, some digesta samples were too small to analyze for energy or nitrogen. However, the basal diet contained twice as much chromic oxide as the treatments since it was not diluted with meat and bone meal. This may have affected the calculations for digesta ME values.

There were differences in the average ME values among meat and bone meal products (Table 5). This is undoubtedly due to differences in nutrient composition,

Table 3: Mean Metabolizable Energy Values for each Assay Method of each Meat and Bone Meal Product (kcal/kg)

Sample	mbm-2		mbm-3		mbm-5		mbm-7	
	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]
Rooster TME_n	2,240[bc]	90	2,469[a]	74	3,026[a]	61	3,329[a]	131
Turkey TMEn	2,528[ab]	81	2,517[a]	74	2,600[bc]	79	3,103[ab]	131
Chick Digesta AMEn	2,135[c]	81	2,436[a]	74	2,555[c]	61	2,705[b]	131
Chick Excreta AMEn	2,508[ab]	81	2,577[a]	74	2,751[abc]	61	3,038[ab]	131
Chick Excreta aAMEn	2,475[abc]	81	2,614[a]	74	2,786[abc]	61	3,081[ab]	131
Poult Digesta AMEn	2,722[a]	90	2,454[a]	74	2,882[ab]	69	2,863[ab]	131
Poult Excreta AMEn	2,586[ab]	90	2,510[a]	74	2,975[a]	61	2,888[ab]	131
Poult Excreta aAMEn	2,611[ab]	90	2,534[a]	74	3,004[a]	61	3,103[ab]	131
Significance	0.0007		NS		<0.0001		0.0579	
	mbm-8		mbm-9		mbm-10		mbm-12	
	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]
Rooster TME_n	2,547[a]	86	3,356[a]	97	2,685[a]	118	1,703[b]	78
Turkey TMEn	2,585[a]	86	2,669[b]	97	2,789[a]	152	2,192[a]	78
Chick Digesta AMEn	2,401[a]	97	2,858[b]	86	2,737[a]	118	1,813[b]	78
Chick Excreta AMEn	2,552[a]	86	3,003[ab]	86	2,820[a]	118	2,168[a]	78
Chick Excreta aAMEn	2,594[a]	86	3,040[ab]	86	2,861[a]	118	2,204[a]	78
Poult Digesta AMEn	2,581[a]	97	2,946[ab]	137	2,891[a]	118	1,872[ab]	78
Poult Excreta AMEn	2,503[a]	86	2,822[b]	86	2,791[a]	118	1,975[ab]	78
Poult Excreta aAMEn	2,530[a]	86	2,851[b]	86	2,811[a]	118	1,999[ab]	78
Significance	NS		0.0017		NS		0.0004	
	mbm-13		mbm-14		mbm-15		mbm-16	
	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]	Mean[1]	SE[2]
Rooster TMEn	2,282[a]	107	2,267[a]	109	2,858[ab]	135	2,106[a]	122
Turkey TMEn	2,010[a]	107	2,355[a]	97	2,583[ab]	120	1,854[a]	136
Chick Digesta AMEn	2,385[a]	107	1,953[a]	97	2,474[b]	120	1,945[a]	122
Chick Excreta AMEn	2,013[a]	107	2,332[a]	97	3,079[a]	120	1,588[a]	122
Chick Excreta aAMEn	2,052[a]	107	2,369[a]	97	3,123[a]	120	1,623a	122
Poult Digesta AMEn	2,330[a]	107	2,067[a]	97	2,785[ab]	135	2,019[a]	122
Poult Excreta AMEn	2,115[a]	107	2,325[a]	97	2,932[ab]	120	2,017[a]	136
Poult Excreta aAMEn	2,137[a]	107	2,355[a]	97	2,974[ab]	120	2,045[a]	136
Significance	NS		NS		0.0062		NS	

[1]Means with no common letter are significantly different. [2]Pooled std error differs due to unequal number of experimental units

reinforcing the variability of the feedstuff (Table 2). The rendered product can vary due to the raw material used and processing techniques employed, such as differences in cooking time, pressure, or temperature (Parsons *et al.*, 1997).

One of the important goals of these experiments was to find differences among chickens and turkeys. The data revealed no differences between the pooled ME values of chickens and turkeys (Table 4). This indicated that the values commonly found for chickens, and Leghorn roosters in particular, can be applied to broilers and turkeys as well. Dale and Fuller (1980) found a similar agreement among roosters, broilers, and turkeys.

The ME_n values from this trial were slightly higher (2,522 kcal/kg) than the National Research Council's (1994) suggestion of 2,495 kcal/kg. Dale (1997) found an ME value for beef meat and bone meal of 2,449 kcal/kg and 2,847 kcal/kg for pork based meat and bone meal. Wang and Parsons (1998) determined a high quality meat and bone meal to have a TME_n of 3,328 kcal/kg and a low quality meat and bone meal to have a TME_n of 2,874 kcal/kg. Others have found results similar to this trial, (Martosiswoyo and Jensen, 1988) indicating the National Research Council's recommendation is too low.

The development of a prediction equation based on proximate and mineral analysis proved disappointing. Carbohydrate and protein content provided the only significant variables. With an R^2 value of 0.42, the equation in Figure 1 does not explain half of the sample variation:

$TME_n = 240.8 - 75.9*(CHO) + 47.8*(CP)$ ($R^2 = 0.42$)

Using gross energy greatly improved the accuracy of the prediction equations (Fig. 2):

Table 4: Mean Metabolizable Energy Comparisons for Meat and Bone Meal Products (kcal/kg)

System	Mean[1]	SE[2]	Significance
ME[3]	2,511[a]	24	NS
TME[3]	2,487[a]	42	
Collection	Mean[1]	SE[2]	Significance
Digesta[4]	2,409[b]	41	0.011
Excreta[4]	2,560[a]	29	
Total[4]	2,487[ab]	42	
Species	Mean[1]	SE[2]	Significance
Chicken	2,501[a]	29	NS
Turkey	2,509[a]	30	

[1]Means with no common letter are significantly different. [2]Standard error differs due to unequal number of experimental units. [3]ME System refers to battery reared birds and TME System refers to tube fed birds. [4]Digesta and excreta samples were collected from battery reared birds and total samples were collected from tube fed birds.

Table 5: Mean Metabolizable Energy Values for each Meat and Bone Meal Product (kcal/kg)

MBM Sample	Mean[1,3]	SE[2]
2	2,518[c]	47
3	2,514[c]	46
5	2,781[ab]	47
7	2,981[a]	46
8	2,503[c]	46
9	2,876[ab]	47
10	2,758[b]	47
12	1,984[ef]	46
13	2,165[de]	46
14	2,245[d]	47
15	2,827[ab]	47
16	1,894[f]	48
Significance	<0.0001	

[1]Means with no common letter are significantly different. [2]Standard error differs due to unequal number of experimental units. [3]Mean is of all replicates of all methods for each sample.

TME_n = -978 - 59.3*(CHO) + 0.9*(GE) (R^2 = 0.77)

Protein quality may be a factor. Since meat and bone meal is high in protein, a large portion of the available energy comes from protein. If the protein content of the product is not readily digested, the available energy will be less than that of a product with higher quality protein. Therefore, two products may have the same amount of protein, but differing amounts of available energy. This problem complicates the development of a useful prediction equation.

It is not surprising that the GE improved predictions since GE is the basis of the TME_n calculation. Unfortunately, proximate analysis does not determine gross energy. Relatively few laboratories have the equipment to determine energy, making it an impractical component for nutritionists to use. When left with only the proximate analysis, the products appear too variable to accurately predict the TME_n.

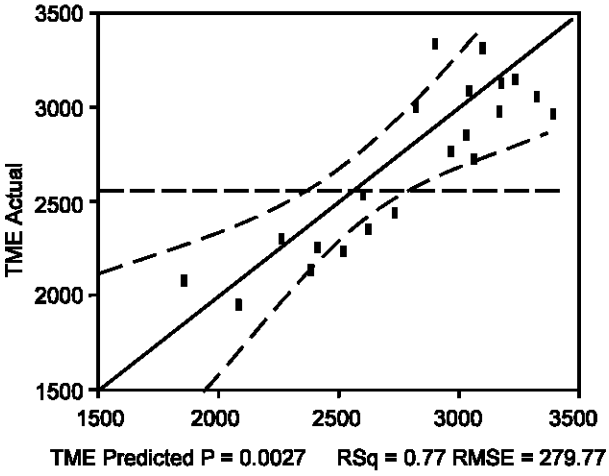

Fig. 2: Best-Fit Prediction Equation of TME_n Value of Meat and Bone Meal from Proximate Analysis and Gross Energy
TME_n = -978 - 59.3*(CHO) + 0.9*(GE)

These equations suggest that it may be worthwhile for nutritionists to invest in bomb calorimetry equipment. The determination of the gross energy of a feedstuff is relatively rapid, taking only a few minutes per sample. The gross energy of the meat and bone meal product, along with proximate and mineral composition values, can be used to calculate the TME_n of the product. This still eliminates the need to conduct animal assays to find the TME_n value of meat and bone meal products.

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