Application of Threonine-Biomass in Broiler Nutrition

Abstract

Threonine is an important amino acid whose effects extend from protein synthesis to immune function, energy metabolism, and nutrient absorption. L-threonine-biomass has a concentration of 80% of Threonine free and additional content of biomass; a source of Threonine, rich in other amino acids, organic acids, vitamins, minerals, and carbohydrates.

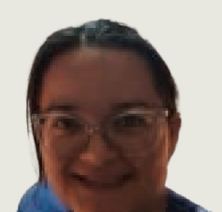
This research was realized with the objective of evaluating the bioequivalence of L-threonine-biomass 80% relative to L-threonine 98.5% in broiler chickens. The trial was performed to evaluate the bioequivalence of L-threonine-biomass 80% based on the slope ratio technique. The experimental diets confirmed that threonine was the limiting amino acid for broilers ($P<0.05$). Weight gain responses were fitted using linear models and, based on slope ratio (b1), [b1 (0.05002) of L-threonine-biomass 80% ÷ b1 (0.05004) of L-threonine 98.5 % × 100 = 99.96%], showed that sources of L-threonine are bioequivalent in broiler nutrition.

The second trial was performed to confirm the bioequivalence between the sources of L-Threonine (L-threonine-biomass 80% versus L-threonine 98.5%), using three diets, one deficient in threonine and two diets were formulated to meet the nutrient requirements of broilers from 1 to 21 days of age, using L-threonine-biomass 80% or L-threonine 98.5% in the diets. The results showed that the threonine deficiency limited the responses of the broilers and, when supplemented with the two sources of threonine, the broilers recovered the performance equally between both.

L-threonine-biomass 80% and L-threonine 98.5% are bioequivalent in broiler chickens

Erikson Raimundo[1]

Michele Lima[2]



Edney Silva[3]*

*This article is based on the research carried out by the Animal Science Department of UNESP, Brazil. The data presented is part of the Doctorate Thesis of Student Mr. Erikson Raimundo and, are not published yet, this article only introduces the key results.

*Corresponding author

[1] PhD Animal Sciences Student, São Paulo State University, Jaboticabal
[2] Brazil University, Department of Veterinary Medicine, Campus of Fernandópolis
[3] São Paulo State University, Department of Animal Sciences, Campus of Jaboticabal

Background

Advances in the production of amino acids on an industrial scale and based on the concept of the ideal protein (Baker and Han, 1994) have enabled more efficient diet formulations, whether in the use of nitrogen and dietary energy, by reducing the energy cost of excreting excess nitrogen in the form of uric acid (Oliveira Neto and Oliveira, 2009).

The α-amino-β-hydroxybutyric acid known as threonine (Thr), is an essential amino acid in animal diets (Kidd and Kerr, 1996). In diets formulated with corn and soybean meal, Thr is the third limiting amino acid for broilers (Warnick and Anderson, 1968; Kidd and Kerr, 1996). Currently, its action on the modulation of cellular metabolism extends from enterocyte integrity to energy metabolism (Corzo et al., 2004; Chen et al., 2017; Tang et al., 2021). However, despite its importance, a unique source of Thr 98,5% is available as in feed-grade market for broilers production.

Studies performed in the last three years showed that L-threonine-biomass, initially with 75% of Thr and additional biomass was similar to L-threonine 98.5% in maintaining broiler performance (Wensley et al., 2020; Lee et al., 2022).

Currently, biotechnological processes have become more efficient, making it possible to increase the Thr-biomass concentration to 80%. This research was carried out with the objective of determining the bioavailability of L-threonine-biomass 80% relative to L-threonine 98.5% for broiler chicken from 1 to 21 days of age.

Application of Threonine-Biomass in Broiler Nutrition*

Methods

Two studies were performed using Cobb® at the experimental facility of the São Paulo State University (UNESP, Jaboticabal city – State of São Paulo). All procedures were approved by Ethics Committee for the Use of Animals (CEUA) under under the Protocol number 0033760/23.

Research 1: Bioavailability of L-Threonine-biomass 80% relative to L-Threonine 98.5%

Birds and experimental design

One thousand five hundred Cobb® chickens with day-1 were housed in the experimental facilities

Each experimental unit was equipped with wood shavings, an infrared lamp for heating the birds; a tubular feeder, and four nipple drinkers.

The completely randomized design was used as experimental design with five treatments, and ten replicates of thirty birds each, in total 50 experimental units.

Bird management

The chicks were housed in a barn with controlled temperature and humidity.

The chicks were weighed individually, and based on body weight, they were distributed with an average body weight of 40 ± 0.2 g. The research lasted 21 days. The daily management was performed according to the Cobb Broiler Management Guide.

Experimental diets and measurements[1]

Treatment 1
BD[2](0.596% Thr, without L-thr sources)
Treatment 2
BD+Thr-Biomass 80%(0.681%Thr)
Treatment 3
BD+Thr-Biomass 80%(0.766%Thr)
Treatment4
BD+L-Thr 98.5%(0.681% Thr)
Treatment5
BD+L-Thr 98.5%(0.766% Thr)

Statistical analysis

The data were analysed to the assumptions of analysis of variance and normality of errors. The F test was used for the ANOVA and when rejected ($P \leq 0.05$), the data were analyzed using a mixed model, considering the treatments as a fixed effect and the experimental unit as a random effect.
The relative bioavailability of L-thr-biomass 80% to L-thr 98.5% was calculated using the slope ratio described by Littell et al. (1997), according to the model: $\xi = \alpha + \beta s Xs + \beta t Xt \pm \check{e}$, where ξ is weight gain; α is intercept; $\check{e}$ is a random error; βs and βt are the slopes for L-threonine 98.5% and 80%, respectively; Xs and Xt are the dietary L-threonine concentrations, respectively

Research 2: Evaluation of L-threonine biomass 80% and L-threonine 98.5% for broilers

Birds and experimental design

Nine hundred Cobb® chickens with one day of age were housed in the experimental facilities.
The experimental unit was equipped with wood savings, an infrared lamp for heating the birds; a tubular feeder, and four nipple drinkers.
The experimental design used was completely randomized design with three treatments, and ten repetitions of thirty birds, totaling thirty experimental units.

Bird management

The chicks were housed in a barn with controlled temperature and humidity.

The chicks were weighed individually, and based on body weight, they were distributed with an average body weight of 40.5 ± 0.1 g. The research lasted 21 days. The daily management was performed according to the Cobb Broiler Management Guide.

Experimental diets and measurements

Treatment 1
BD[3](0.596% thr, without L-thr sources)
Treatment 2
BD+L-Thr-Biomass 80%(0.852%thr)
Treatment 3
BD+L-Thr-Biomass 98.5%(0.852%thr)

Statistical analysis

The data were analysed to the assumptions of analysis of variance and normality of errors. The F test was used for the ANOVA and when rejected ($P \leq 0.05$), the multiple comparison test was applied, using the Tukey test with a significance of 5%. Statistical analyses and estimation of model parameters were performed using SAS 9.4 (Statistical Analysis for Windows, SAS Institute Inc., Cary, NC, USA).

[1]The experimental levels of Thr 0.596%, 0.681%, and 0.766% were defined as proposed by Littel et al. (1997).
[2]The basal diet (BD) was formulated without a source of L-Thr and the other levels (0.681% and 0.766%) were obtained with the supplementation of the respective evaluated sources (L-Threonine-biomass 80% and L-Threonine 98.5%). It was considered 100% as the digestibility coefficient for L-Threonine 98.5%.
[3]BD - Basal diet [4]WG - Weight gain

Trial Results

The assumptions for analysis of variance were tested and met. The results of the analysis of variance for weight gain, in the period from 1 to 21 days of age, are presented in Table 1. According to the average values obtained from the treatments, the reduction in weight gain obtained with BD (0.596%) was approximately 9.2% and 15.0% in relation to the first (0.681%) and second (0.766%) level of threonine in the diet, regardless of the source of threonine.

The bioavailability of L-Thr-biomass 80% based on dietary supplementation of L-Thr-biomass 80% and L-Thr 98.5% were estimated based on weight gain in broiler chickens.

Weight gain response was adjusted using the linear model as a function of supplemental L-threonine intake corrected for Thr concentration in each source (Figure 1). When analyzing the intercept hypothesis, the estimated difference for the basal diet was -0.282 ($P= 0.334$). No significant difference was observed between the evaluated sources of Thr ($P= 0.832$), with a single intercept being used for both adjusted lines. The equations obtained were the following:

- **Threonine-biomass 80%: WG[4] = 0.9672** + 0.05002** × Thr Intake**
  (**$P \leq 0.01$)
- **Threonine 98.5%: WG = 0.9672** + 0.05004** × Thr Intake**
  (**$P \leq 0.01$)

Based on the slope ratio (b1), the sources were compared b1 (0.05002) of L-threonine-biomass 80% ÷ b1 (0.05004) of L-threonine 98.5% × 100, resulting in a value of 99.96% relative bioavailability (RBA).

At the Table 2 and Figure 2 shows the weight gain results of broiler chickens submitted to three diets. Basal diet, without L-Thr supplementation, formulated to meet 0.596% of Thr and, two diets to meet the requirement of 0.852% of threonine during the phase from 1 to 21 days and supplemented with L-Thr-biomass 80% and L-Thr 98.5%. It was observed that both sources of L-Threonine improve the weight gain ($P<0.001$).

Application of Threonine-Biomass in Broiler Nutrition

Trial Results

Table 1. Threonine intake and weight gain of broilers from 1 to 21 days of age.

		Thr in the Diet (%)	L-Thr In the Diet (g/kg)	Intake of L-Thr (g/bird)	Weight gain (kg/bird)
Treatments	T1: Basal	0.596	0	0	0.937
	T2: Basal+Thr80%	0.681	1.057	1.113	1.023
	T3: Basal+Thr80%	0.766	0.863	2.256	1.078
	T4: Basal+L-Thr98.5%	0.681	0.863	1.136	1.024
	T5: Basal+L-Thr98.5%	0.766	1.731	2.319	1.082
Average				1.364	1.028
Stderr				0.04	0.04
P-value				<0.0001	<0.0001
Coefficient of variation, %				3.08	4.71

[1]BTPS: Brazilian Tables for Poultry and Swine

Table 2. Effect of L-threonine on the weight gain of broilers at 21 days

		Level of Thr in the Diet (%)*	Supplementary L-Thr in Diet (kg/ton)	Weight gain (kg)
Treatments	T1: Basal	0.596	0	0.937[b]
	T2: Basal+L-Thr80%	0.852	3.200	1.096[a]
	T3: Basal+L-Thr98.5%	0.852	2.595	1.076[a]
Average				
Stderr				0.04
P-value				<0.0001
Coefficient of variation, %				4.05

* Recommendation of Rostagno et al. (2017) – Brazilian Table.

Figure 1. Bioavailability of L-Threonine-biomass 80% compared to L-Thr 98.5% for weight gain based on linear slope ratio model.

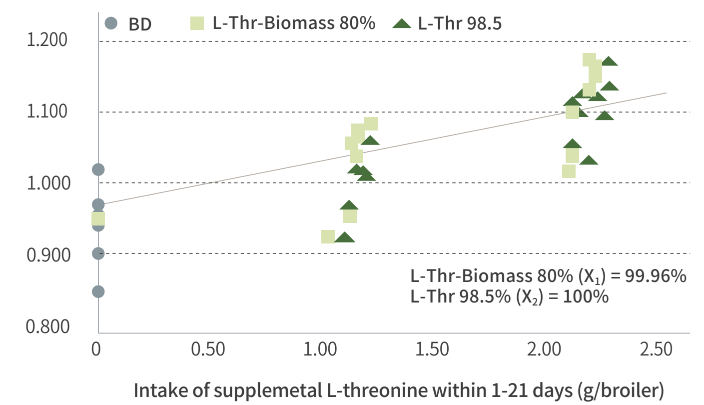
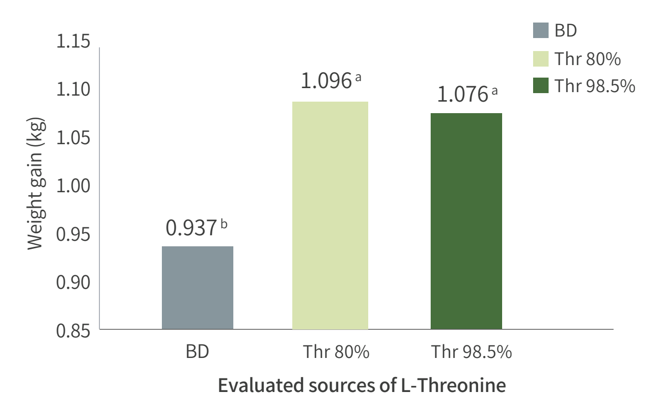


Figure 2. Responses of broilers to sources of L-Threonine (P>0.05).



Application of Threonine-Biomass in Broiler Nutrition

Discussion

This research aimed to evaluate the bioequivalence between sources of Thr, L-threonine-biomass 80% versus L-threonine 98.5%, for broiler chickens. Threonine levels used affected birds weight gain response (Table 1). The slope ratio (0.05002 ÷0.05004×100), resulted in RBA of 99.96% of L-threonine- biomass 80%.

Previous studies (Wensley et al., 2020) evaluated the performance responses of broilers fed L-threonine-biomass 75% and L-threonine 98% and, applying analysis of variance the authors found no difference between treatments. Despite its validity for interpreting the response, this type of experimental design is based on multiple comparisons of means and does not support inference based on the unit response rate. Therefore, the applied method is complementary to the results of Wensley et al. (2020), seems to be the first study to determine the bioequivalence of L-threonine-biomass 80%.

The second assay confirmed that the sources of Thr were similar (Figure 2). Despite the similarity with the experimental design used by Wensley et al. (2020), the experimental levels used in this research reduced the responses of birds fed with the basal diet, on the other hand, with the L-Thr sources an increase in weight gain was found giving + 14.8% to L-Threonine 98.5% and + 17.0% to L-threonine-biomass 80%. The result found in this trial eliminates any hypothesis about an antinutritional effect present in the biomass.

Conclusion

L-threonine-biomass 80% and L-threonine 98.5% are bioequivalent for broilers. Also, broilers fed with a diet to meet 0.852% of SID threonine with both sources of L-Threonine presented the same weight gain.

References

· Baker, D. H., and Y. Han. 1994. Ideal amino acid profile for chicks during the first three weeks posthatching. Poult. Sci. 73:1441–1447.

· Chen, Y. P., Y. F. Cheng, X. H. Li, W. L. Yang, C. Wen, S. Zhuang, and Y. M. Zhou. 2017. Effects of threonine supplementation on the growth performance, immunity, oxidative status, intestinal integrity, and barrier function of broilers at the early age. Poult. Sci. 96:405–413.

· Corzo, A., M. T. Kidd, D. J. Burnham, and B. J. Kerr. 2004. Dietary glycine needs of broiler chicks. Poult. Sci. 83:1382–1384.

· Hermann, T. 2003. Industrial production of amino acids by coryneform bacteria. J. Biotechnol. 104:155–172.

· Kidd, M. T., and B. J. Kerr. 1996. L-Threonine for poultry: A review. J. Appl. Poult. Res. 5:358–367

· Lee, D. T., J. T. Lee, A. J. Ashworth, M. T. Kidd, A. Mauromoustakos, and S. J. Rochell. 2022. Evaluation of a threonine fermentation product as a digestible threonine source in broilers. J. Appl. Poult. Res. 31:100252.

· Littell, R. C., P. R. Henry, A. J. Lewis, and C. B. Ammerman. 1997. Estimation of Relative Bioavailability of Nutrients Using SAS Procedures. J. Anim. Sci. 75:2672–2683.

· Oliveira Neto, A. R. de, and W. P. de Oliveira. 2009. Aminoácidos para frangos de corte. Rev. Bras. Zootec. 38:205–208.

· Tang, Q., P. Tan, N. Ma, and X. Ma. 2021. Physiological functions of threonine in animals: Beyond nutrition metabolism. Nutrients 13.

· Warnick, R. E., and J. O. Anderson. 1968. Limiting essential amino acids in soybean meal for growing chickens and the effects of heat upon availability of the essential amino acids. Poult. Sci. 47:281–287.

· Wensley, M. R., J. C. Woodworth, J. M. DeRouchey, S. S. Dritz, M. D. Tokach, R. D. Goodband, H. G. Walters, B. A. Leopold, C. D. Coufal, K. D. Haydon, and J. T. Lee. 2020. Effects of amino acid biomass or feed-grade amino acids on growth performance of growing swine and poultry. Transl. Anim. Sci. 4:49–58.