



Digestibility of animal ingredients for tambaqui and Nile tilapia

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Digestibilidade de ingredientes de origem animal para tambaqui e tilápia do Nilo

RESUMO

Objetivo – Determinar a digestibilidade proteica, energética e de fósforo das farinhas de carne e ossos, peixe, sangue, penas e vísceras aplicados em rações para tambaqui (*Colossoma macropomum*) e tilápia do Nilo (*Oreochromis niloticus*).

Metodologia – Foram utilizados 48 tambaquís e 48 tilápias com peso médio $198,15 \pm 3,11$ e $134,89 \pm 2,22$ gramas, respectivamente. O experimento foi montado em delineamento inteiramente casualizado, esquema fatorial (grupo x espécie), composto por 10 tratamentos (cinco alimentos x duas espécies), três repetições no tempo e oito peixes por unidade experimental.

Originalidade/Relevância – O estudo comparou, de forma simultânea, a digestibilidade de cinco farinhas de origem animal por duas das principais espécies da aquicultura brasileira, tambaqui e tilápia, sob um mesmo protocolo experimental. Essa abordagem forneceu dados padronizados e comparáveis, pouco comuns na literatura. A relevância prática é notória, já que os coeficientes de digestibilidade obtidos podem ser diretamente utilizados na formulação de rações, contribuindo para a redução de custos, melhoria da eficiência alimentar e simplificação da formulação para múltiplas espécies.

Resultados – O tambaqui obteve maior coeficiente de digestibilidade aparente (CDA) para proteína bruta (PB) da farinha de peixe (87,20%) e a tilápia apresentou melhor CDA para PB das farinhas: carne e ossos (85,43%), peixe (86,38%) e vísceras (85,92%). Quanto ao coeficiente de digestibilidade aparente da energia bruta (CDAEB), a tilápia obteve melhor resultado com a farinha de carne e ossos (89,69%) e o tambaqui demonstrou maior CDAEB para as farinhas: carne e ossos (82,77%), peixe (85,01%) e vísceras (86,04%). Ambas as espécies apresentaram maior CDA do fósforo total (FT) para a farinha de sangue. As espécies aproveitaram de forma semelhante às farinhas de peixe (68,42%), sangue (90,36%), penas (55,68%) e vísceras (60,01%). Em termos práticos e considerando as excepcionalidades, podem-se adotar valores médios para os coeficientes de digestibilidade de alimentos de origem animal para ambas as espécies, visto que, as diferenças observadas para o CDA da PB para os dois peixes foram apenas para as farinhas de sangue e vísceras.

Contribuições Teóricas/Metodológicas – A pesquisa atualiza os coeficientes de digestibilidade de proteína, energia e fósforo de ingredientes de origem animal para peixes tropicais, demonstrando que, com exceções, valores médios podem ser utilizados para tambaqui e tilápia, facilitando a formulação de rações em policultivos. O uso de delineamento fatorial com duas espécies e cinco ingredientes, aliado a três repetições no tempo, confere maior robustez estatística e confiabilidade aos resultados.

Contribuições Sociais e Ambientais – O estudo contribui para a eficiência alimentar e redução de custos na piscicultura, beneficiando pequenos e médios produtores com maior previsibilidade zootécnica e melhor planejamento produtivo. Além disso, favorece a sustentabilidade ao reduzir a carga de nutrientes no ambiente e estimular o uso de coprodutos da indústria animal, promovendo a economia circular no setor.

PALAVRAS-CHAVE: *Colossoma macropomum*. Farinha de origem animal. *Oreochromis niloticus*. Valor nutricional.

Digestibility of animal ingredients for tambaqui and Nile tilapia

ABSTRACT

Objective – This study aimed to determine the protein, energy, and phosphorus digestibility of animal ingredients (meat and bone, fish, blood, feather, and viscera meals) in diets for tambaqui (*Colossoma macropomum*) and Nile tilapia (*Oreochromis niloticus*).

Methodology – A total of 48 tambaqui and 48 Nile tilapia were used, with a mean weight of 198.15 ± 3.11 g and 134.89 ± 2.22 g, respectively. The experiment followed a completely randomized design with a factorial arrangement (ingredient × species), comprising 10 treatments (five ingredients × two species), 3 temporal replications, and 8 fish per experimental unit.

Originality/Relevance – This study simultaneously compared the digestibility of five animal meal ingredients for two key species of Brazilian aquaculture, namely tambaqui and tilapia, under a combined experimental protocol. This approach yielded standardized and directly comparable data, which are scarce in the literature. The practical

relevance is high, as the digestibility coefficients obtained can be directly applied in feed formulation, contributing to cost reduction, improved feed efficiency, and simplified diet formulation for multispecies systems.

Results – Tambaqui showed a higher apparent digestibility coefficient (ADC) for crude protein in fish meal (87.20%), whereas tilapia achieved higher ADCs for crude protein in meat and bone meal (85.43%), fish meal (86.38%), and viscera meal (85.92%). Regarding the ADC of gross energy, tilapia performed best with meat and bone meal (89.69%), whereas tambaqui exhibited higher ADC values for meat and bone (82.77%), fish (85.01%), and viscera (86.04%) meals. Both species showed the highest ADC of total phosphorus in blood meal. Furthermore, the species exhibited similar phosphorus ADC values for fish meal (68.42%), blood meal (90.36%), feather meal (55.68%), and viscera meal (60.01%). In practical terms and considering a few exceptions, the same digestibility coefficients of animal meal ingredients can be adopted for both species, given that significant differences were observed only in crude protein digestibility for blood and viscera meals.

Theoretical/Methodological Contributions – This study provided updated digestibility coefficients for crude protein, gross energy, and phosphorus in animal meal ingredients used in diets for tropical fish. It demonstrated that, with few exceptions, mean values can be applied to both tambaqui and tilapia, thereby simplifying feed formulation in polyculture systems. The factorial design adopted herein, with two species × five ingredients and three temporal replications, ensured the statistical robustness and reliability of the results.

Social and Environmental Contributions – The research contributes to feed efficiency and cost reduction in aquaculture, benefiting small- and medium-scale farmers by improving production planning and predictability. Furthermore, it supports environmental sustainability by reducing nutrient discharge and encouraging the utilization of animal industry co-products, fostering a circular economy in the aquaculture sector.

KEYWORDS: *Colossoma macropomum*. Animal meal. *Oreochromis niloticus*. Nutritional value.

Digestibilidad de ingredientes de origen animal para tambaquí y tilapia del Nilo

RESUMEN

Objetivo – Esta investigación tuvo como objetivo determinar la digestibilidad de proteínas, energía y fósforo de ingredientes de origen animal (harinas de carne y hueso, pescado, sangre, plumas y vísceras) en dietas para tambaquí (*Colossoma macropomum*) y tilapia del Nilo (*Oreochromis niloticus*).

Metodología – Se utilizaron 48 tambaquíes y 48 tilapias del Nilo, con un peso promedio de $198,15 \pm 3,11$ g y $134,89 \pm 2,22$ g, respectivamente. El experimento siguió un diseño completamente aleatorizado con un arreglo factorial (ingrediente × especie), compuesto por 10 tratamientos (cinco ingredientes × dos especies), tres repeticiones temporales y ocho peces por unidad experimental.

Originalidad/Relevancia – Esta investigación comparó simultáneamente la digestibilidad de cinco ingredientes de harinas animales en dos especies clave de la acuicultura brasileña, el tambaquí y la tilapia, bajo un protocolo experimental combinado. Este enfoque permitió obtener datos estandarizados y directamente comparables, que son escasos en la literatura. La relevancia práctica es alta, ya que los coeficientes de digestibilidad obtenidos pueden aplicarse directamente en la formulación de raciones, contribuyendo a reducir costos, mejorar la eficiencia alimenticia y simplificar la formulación en sistemas con múltiples especies.

Resultados – El tambaquí mostró un coeficiente de digestibilidad aparente (CDA) más alto para la proteína bruta en la harina de pescado (87,20%), mientras que la tilapia alcanzó mayores CDA de proteína bruta en las harinas de carne y hueso (85,43%), pescado (86,38%) y vísceras (85,92%). En cuanto al CDA de la energía bruta, la tilapia presentó el mejor rendimiento con harina de carne y hueso (89,69%), mientras que el tambaquí exhibió valores más altos de CDA para las harinas de carne y hueso (82,77%), pescado (85,01%) y vísceras (86,04%). Ambas especies mostraron la mayor CDA de fósforo total en la harina de sangre. Se observaron valores similares de digestibilidad del fósforo para las harinas de pescado (68,42%), sangre (90,36%), plumas (55,68%) y vísceras (60,01%). En términos prácticos, y salvo algunas excepciones, se pueden adoptar los mismos coeficientes de digestibilidad para los ingredientes de harinas animales en ambas especies, dado que solo se observaron diferencias significativas en la digestibilidad de proteína bruta en las harinas de sangre y vísceras.

Contribuciones Teóricas y Metodológicas – Esta investigación proporcionó coeficientes actualizados de digestibilidad de proteína, energía y fósforo en ingredientes de harinas animales utilizados en dietas para peces tropicales. Se demostró que, con pocas excepciones, los valores promedio pueden aplicarse tanto al tambaquí como



a la tilapia, simplificando así la formulación de alimentos en sistemas de policultivo. El diseño factorial adoptado, con dos especies $\times$ cinco ingredientes y tres repeticiones temporales, fortaleció la robustez estadística y la confiabilidad de los resultados.

Contribuciones Sociales y Ambientales – La investigación contribuye a mejorar la eficiencia alimenticia y reducir los costos en la acuicultura, beneficiando a productores pequeños y medianos mediante una mejor planificación y previsibilidad de la producción. Además, promueve la sostenibilidad ambiental al disminuir la descarga de nutrientes y fomentar el aprovechamiento de subproductos de la industria animal, impulsando una economía circular en el sector acuícola.

PALABRAS CLAVE: *Colossoma macropomum*. Harina de origen animal. *Oreochromis niloticus*. Valor nutricional.

1 INTRODUCTION

Brazilian aquaculture includes several prominent species of fish. Among them, Nile tilapia (*Oreochromis niloticus*) and tambaqui (*Colossoma macropomum*) are notable for their tolerance to low-oxygen environments, rapid growth, consistent fry production, strong market acceptance, and high nutritional value (SENA, 2012). Native to Africa, Nile tilapia is characterized by an omnivorous feeding habit and an easy acceptance of feed from the post-larval stage onward. Globally, tilapia aquaculture represents one of the most important sources of fish. The species is now present in more than 140 countries across five continents, with a steadily increasing global production. In 2020, approximately 6,100,719 t of tilapia entered the market. In Brazil alone, production reached 361,286.198 t in 2021 (FAO, 2018; FAO, 2020; IBGE, 2022).

Tambaqui is a characid fish belonging to the class Actinopterygii and the order Characiformes. It has an omnivorous feeding habit and is native to the Amazon and Orinoco River basins (QUIRINO *et al.*, 2024; SILVA *et al.*, 2024). This species reaches sexual maturity between the third and fourth year of age and, in natural environments, can grow up to 1 m in length and 45 kg in weight (VARELA *et al.*, 2021). In 2022, tambaqui production, together with other native species, exceeded 267,000 t, representing 31.04% of the total fish production in Brazil (PEIXE BR, 2023).

With the growing cultivation of these species, particularly tambaqui, researchers have intensified studies aimed at improving nutrition under confinement conditions (PORTO *et al.*, 2020). Given that commercial feed represents the main production cost in intensive systems and is the primary nutrient source for confined animals, optimizing feed formulations is essential in aquaculture (SENA, 2012; BASSANI; ROCHA, 2020).

The inclusion of animal by-products in fish feed contributes to the sustainability of the sector by reducing solid waste generation, greenhouse gas emissions, and environmental impacts, such as eutrophication (AGUILAR; PAULINO, 2024). Utilizing these ingredients, which would otherwise be destined for landfills or incineration, aligns with circular economy principles and reduces pressure on conventional protein sources, such as fish meal. Moreover, this approach promotes more efficient nutrition, with reduced excretion of nutrients such as nitrogen and phosphorus, which are associated with water quality degradation in nurseries and water bodies (RESENDE; CUSTÓDIO, 2024; SILVA; SANO; CHAVES, 2024). This approach can also represent an economically viable alternative by lowering production costs.

To achieve these benefits and enhance production efficiency, it is essential to determine the nutritional requirements and digestibility coefficients of feed ingredients (of both animal and plant origin) in order to develop balanced diets that improve animal performance and minimize waste generation (PORTO *et al.*, 2020). Fish diets typically incorporate ingredients derived from agricultural and animal by-products (ARAGÃO *et al.*, 2022). The latter give rise to animal meals, which are rich in amino acids, energy, fat, and minerals compared with plant-based meals. However, the inclusion of these meals must be carefully balanced to prevent undesirable effects such as rancidity and nutritional imbalances (MACHADO; DILELIS; LIMA, 2021; MENEZES *et al.*, 2021; SILVA, E. *et al.*, 2024).

Among the minerals present in animal meals, phosphorus stands out because it is

fully available for digestion (not bound to phytate). Another important phosphorus source is dicalcium phosphate, the primary inorganic supplement used in feed, particularly those formulated with plant-based ingredients (MEYER *et al.*, 2019). However, studies with non-ruminants indicated that the digestibility coefficient of these sources is less than 100% (MEYER *et al.*, 2019; ROSTAGNO *et al.*, 2017).

As feed formulation must consider the fractions of nutrients and energy available for animal metabolism, determining the digestibility coefficients of the nutritional components of feed ingredients is critical to developing balanced diets that meet fish nutritional requirements, as demonstrated by Aragão *et al.* (2020) and Rodrigues *et al.* (2022). Considering the importance of tambaqui and Nile tilapia in fish farming, this study aimed to determine the proximate composition and the protein, energy, and phosphorus digestibility of animal meal ingredients for these species.

2 MATERIAL AND METHODS

The experiment was conducted in accordance with ethical standards for animal research and was approved by the Animal Research Ethics Committee at the Federal University of Maranhão (CEUA-UFMA, protocol No. 23115.012035/2018-37). The study took place at the Laboratory of Nutrition and Feeding of Aquatic Organisms of Maranhão (LANUMA), located at the Chapadinha Science Center of UFMA, in the municipality of Chapadinha, Maranhão State, Brazil.

Five animal ingredients were evaluated: fish meal, meat and bone meal, viscera meal, feather meal, and blood meal. For determination of the apparent digestibility coefficient (ADC) for crude energy, crude protein, and total phosphorus in each ingredient, 48 tambaqui (*C. macropomum*) and 48 Nile tilapia (*O. niloticus*) were used. All fish were in the grower phase. The mean weights were 198.15 ± 3.11 g for tambaqui and 134.89 ± 2.22 g for Nile tilapia.

The experiment followed a completely randomized factorial design with 10 treatments (5 ingredients $\times$ 2 species), 3 temporal repetitions (i.e., the same set of treatments repeated at 3 different time points), and 8 fish per experimental unit. At each repetition, collections continued until sufficient quantities of sample were obtained for laboratory analyses.

Six diets were prepared, including a reference diet based on corn and soybean meal (Table 1) and five experimental diets, each containing 69.9% of the reference diet and 30% of the test ingredient. ADCs were determined by the indirect method (NRC, 2011). For this, 0.10% chromium oxide (Cr_2O_3) was added to the experimental diets as an indigestible external marker, resulting in a composition of 99.90% experimental diet and 0.10% Cr_2O_3 . Additionally, a separate group of eight fish per species was used to determine the digestibility coefficient of the reference diet (99.90% reference feed + 0.10% external indicator).

Table 1 – Ingredient and chemical composition of the reference diet (as-fed basis)

Ingredient	(%)
Soybean meal (45% crude protein)	54.995
Corn	36.386
Soybean oil	4.400
D,L-Methionine (99%)	0.159
L-Threonine (98.5%)	0.065
Calcitic limestone	0.323
Dicalcium phosphate	2.594
Vitamin and mineral supplement ⁽¹⁾	0.500
Vitamin C ⁽²⁾	0.050
Salt	0.508
Antioxidant (BHT)	0.020
Calculated composition⁽³⁾	
Crude protein (%) (4)	29.09
Gross energy (kcal kg ⁻¹) (4)	4,231.00
Ether extract (%)	6.61
Crude fiber (%)	3.54
Total Ca (%)	0.90
Total P (%) (4)	0.97
Total Na (%)	0.22
Total lysine (%)	1.618
Total Met + Cys (%)	0.982
Total threonine (%)	1.159
Total tryptophan (%)	0.368
Gross energy/crude protein ratio (kcal g ⁻¹)	14.72

Source: prepared by the authors (2023).

⁽¹⁾Commercial vitamin and mineral supplement (5 kg t⁻¹), providing per kilogram of product: vitamin A, 1,200,000 IU; vitamin D₃, 200,000 IU; vitamin E, 1200 mg; vitamin K₃, 2400 mg; vitamin B₁, 4800 mg; vitamin B₂, 4800 mg; vitamin B₆, 4800 mg; vitamin B₁₂, 4800 mg; vitamin C, 48 g; folic acid, 1200 mg; calcium pantothenate, 12,000 mg; biotin, 48 mg; choline chloride, 108 g; niacin, 24,000 mg; Fe, 50,000 mg; Cu, 3000 mg; Mn, 20,000 mg; Zn, 30,000 mg; I, 100 mg; Co, 10 mg; and Se, 100 mg. ⁽²⁾Vitamin C was provided as calcium ascorbate-2-monophosphate, containing 42% active ingredient. ⁽³⁾Composition was calculated based on Rostagno *et al.* (2017). ⁽⁴⁾Nutrient values and digestibility coefficients for Nile tilapia were based on Furuya *et al.* (2010).

Before the beginning of the experiment, the fish were kept in polyethylene tanks (1000 L) equipped with individual water supply and drainage systems, as well as supplementary aeration. During the pre-experimental period, fish were fed the respective experimental diets. Then, in the experimental period, the fish were transferred to conical fiberglass incubators (220 L) adapted for feces collection and equipped with an aeration system. Throughout the day, fish were fed the experimental diets over six meals (8:00, 10:00, 12:00, 14:00, 15:00, and 16:00 h). Feed was provided *ad libitum* by manual broadcasting in successive rounds, thereby allowing maximum intake until apparent satiety. About 30 min after the last feeding, the incubators were cleaned, 70% of the water was replaced, and each incubator was covered with black tarpaulin to prevent contamination from airborne residues.

Polyethylene feces collectors (mean volume of 240 mL, adapted for decantation by the modified Guelph method) were attached to the lower portion of fiberglass incubators, as described by Abimorad and Carneiro (2004). Fecal fermentation was prevented by maintaining the collectors in thermal boxes with ice throughout the collection period. Fecal samples were collected between 18:00 and 06:00 h, at approximately 2 h intervals, totaling six collections per group. During each collection, the valve connecting the collector to the incubator was closed, the collector was removed, and the supernatant was discarded. The decanted feces

were placed in a Petri dish and dried in a forced-air oven at 65 °C for 72 h for laboratory analysis.

Feed ingredients were ground, mixed, moistened with water heated to approximately 50 °C, and pelletized using a meat grinder to minimize nutrient leaching. The pellets were dried in a forced-air oven at 65 °C for 72 h, ground, and sieved through a 3.00 mm mesh sieve to obtain pellets measuring approximately 3 mm in diameter.

The ADC of crude protein, gross energy, and total phosphorus in experimental and reference diets was estimated by calculating the indigestibility factor using the equations described by Pezzato *et al.* (2004) (Equations 1 and 2):

$$ADC = 100 - \left[100 \times \left(\frac{\text{Indicator in diet}}{\text{Indicator in feces}} \right) \times \left(\frac{\text{Nutrient in diet}}{\text{Nutrient in feces}} \right) \right] \quad (1)$$

where ADC (%) is the apparent digestibility coefficient of the diet.

$$ADC_{ing} = \frac{ADC_e - (b \times ADC_r)}{a} \quad (2)$$

where ADC_{ing} is the apparent digestibility coefficient of the ingredient; ADC_e is the apparent digestibility coefficient of the experimental diet; ADC_r is the apparent digestibility coefficient of the reference diet; b is the percentage of the reference diet; and a is the percentage of the test ingredient.

Water temperature was monitored twice daily (08:00 and 16:00 h) using a mercury bulb thermometer graduated from 0 to 50 °C. Water pH, dissolved oxygen, and ammonia (NH_3) levels were measured every 3 days by using a pH meter, an oximeter, and a commercial ammonia test kit, respectively.

The proximate composition of test ingredients and diets was determined by a private laboratory (C.B.O. Análises Laboratoriais Ltda., Valinhos, São Paulo, Brazil), except for dry matter, mineral matter, and chromic oxide, which were determined at the Animal Nutrition Laboratory of the UFMA, following the procedures described by Detmann *et al.* (2021).

Statistical analyses were performed using InfoStat software (2008). Data were subjected to analysis of variance at the 5% significance level. The effect of species on ADCs was assessed using the *F*-test. Significant differences in ADCs were compared using Tukey's test ($p \leq 0.05$).

3 RESULTS

Water physicochemical parameters, both in the tanks and incubators, remained stable throughout the experiment and were mostly within the ranges considered suitable for the cultivation of tambaqui and Nile tilapia. The mean daily temperature was 26.01 ± 1.10 °C. Dissolved oxygen levels were 6.67 ± 1.15 mg L⁻¹, exceeding the recommended 5 mg L⁻¹ threshold. Nevertheless, no labial prolapse was observed in tambaqui (IZEL-SILVA *et al.*, 2020). The pH was 6.33 ± 0.23 , being within the optimal range of 6 to 9 (GOMES; BORGES; FERREIRA, 2025). Ammonia levels remained below 1.00 mg L⁻¹, within acceptable limits (NASCIMENTO, L. T. *et al.*, 2020).

Data on the composition of the tested feed ingredients are presented in Table 2, and the ADCs for crude protein are shown in Table 3. The composition of the tested animal meal ingredients was relatively similar to the values and variations reported in Brazilian tables for

tilapia nutrition (FURUYA *et al.*, 2010), as well as in tables for feed composition and nutritional requirements of poultry and swine (ROSTAGNO *et al.*, 2017). It should be noted that these meals typically do not exhibit a standardized composition due to variations in source materials and processing methods (MACHADO; DILELIS; LIMA, 2021).

Table 2 – Nutritional composition of the feed ingredients analyzed in the experiment (as-fed basis)

Component	Meat and bone meal	Fish meal	Blood meal	Feather meal	Viscera meal
DM (%)	94.81	89.31	93.49	93.22	93.03
CP (%)	49.88	64.34	84.44	80.48	59.00
GE (kcal kg ⁻¹)	3.795	4.365	5.334	5.398	4.800
EE (%)	13.12	8.02	1.66	7.68	15.20
MM (%)	32.27	23.55	3.41	2.59	17.44
TP (%)	5.49	3.29	0.12	0.43	2.70

Source: prepared by the authors (2023).

DM, dry matter; CP, crude protein; GE, gross energy; EE, ether extract; MM, mineral matter; TP, total phosphorus.

The crude protein content of meat and bone meal (49.88%) falls within the range reported in the literature, which varies from 37.9% to 59.0% (PEZZATO *et al.*, 2002; FURUYA *et al.*, 2010; ROSTAGNO *et al.*, 2017). Similarly, gross energy (3795 kcal kg⁻¹) is consistent with previously reported values, which range from 3022.53 to 4176.00 kcal kg⁻¹ (PEZZATO, 2002; ABIMORAD; CARNEIRO, 2004; FURUYA *et al.*, 2010; ROSTAGNO *et al.*, 2017). Total phosphorus (5.49%) is also within the range observed in other studies, which report values from 5% to 7.5% (ROSTAGNO *et al.*, 2017; MACHADO; DILELIS; LIMA, 2021).

In fish meal, crude protein (64.34%) and gross energy (4365 kcal kg⁻¹) contents were higher than those reported in previous studies, which show minimum values of 53.93% for protein and 3436.13 kcal kg⁻¹ for energy and maximum values of 63.80% and 4199 kcal kg⁻¹, respectively (GONÇALVES; CARNEIRO, 2003; ABIMORAD; CARNEIRO, 2004; FURUYA *et al.*, 2010; ROSTAGNO *et al.*, 2017). On the other hand, total phosphorus (3.29%) was lower than the value reported in Brazilian tables for tilapia nutrition (4.33%) (FURUYA *et al.*, 2010).

Blood meal had a crude protein content of 84.44%, which falls within the range reported by various authors (73.44% to 84.50%). Its gross energy (5334 kcal kg⁻¹) was similar to literature values, which range from 4756 to 5725.83 kcal kg⁻¹. By contrast, total phosphorus (0.12%) was lower than reported values (0.24% to 0.32%) (PEZZATO *et al.*, 2002; GONÇALVES; CARNEIRO, 2003; HENN, 2004; FURUYA *et al.*, 2010; PEZZATO *et al.*, 2012; ROSTAGNO *et al.*, 2017; MACHADO, DILELIS; LIMA, 2021).

Regarding feather meal, crude protein (80.48%) and gross energy (5398 kcal kg⁻¹) contents were similar to values reported in Brazilian tables for poultry and swine, which range from 76.20%–83.10% and 5137–5238 kcal kg⁻¹, respectively (ROSTAGNO *et al.*, 2017). The observed values for ether extract (7.68%) and mineral matter (2.59%) were lower than the respective 15.60% and 5.40% reported in a review by Henriques, Rodrigues and Lazzari (2018).

For viscera meal, the values obtained in this study for crude protein (59.00%) and gross energy (4800 kcal kg⁻¹) were similar to those reported by Furuya *et al.* (2010) and Abimorad and Carneiro (2003), which average 58.64% and 4616.71 kcal kg⁻¹, respectively. The total phosphorus content (2.70%) of viscera meal was within the range of 2.54% to 4.68% reported by Rostagno *et al.* (2017). Overall, the observed differences in the chemical

composition of feed ingredients can be attributed to the lack of standardization resulting from variations in raw materials and processing methods (VIEIRA *et al.*, 2007).

For the ADCs of crude protein, a significant interaction ($p < 0.05$) between feed ingredient and species was observed, indicating that crude protein utilization may vary between species (Table 3). No significant differences were found between species for the ADC of crude protein in meat and bone meal, fish meal, or feather meal, with mean values of 84.25%, 86.79%, and 66.04%, respectively, for both species. However, significant differences were observed for blood meal and viscera meal: tilapia showed a higher digestibility of viscera meal (85.92%) than tambaqui (82.68%), whereas tambaqui exhibited a higher digestibility of blood meal (46.72%) than tilapia (43.39%).

Table 3 – Apparent digestibility coefficients of crude protein and digestible protein contents of animal meal ingredients in diets for tambaqui and Nile tilapia

Species	Meat and bone meal	Fish meal	Blood meal	Feather meal	Viscera meal
Apparent digestibility coefficient (%)					
Tambaqui	83.08 ± 0.81 ^{Ab}	87.20 ± 1.27 ^{Aa}	46.72 ± 0.85 ^{Ad}	65.21 ± 0.74 ^{Ac}	82.68 ± 0.34 ^{Bb}
Nile tilapia	85.43 ± 1.40 ^{Aa}	86.38 ± 0.99 ^{Aa}	43.39 ± 1.04 ^{Bc}	66.87 ± 0.93 ^{Ab}	85.92 ± 1.28 ^{Aa}
Digestible crude protein content (%)					
Tambaqui	41.44 ± 0.40	56.10 ± 0.81	39.45 ± 0.84	52.48 ± 0.64	48.78 ± 0.20
Nile tilapia	42.61 ± 0.69	55.58 ± 0.64	36.64 ± 0.98	53.81 ± 0.75	50.69 ± 0.76

Source: prepared by the authors (2023).

Results are presented as mean ± standard deviation. Means followed by the same uppercase letters within columns or the same lowercase letters within rows do not differ significantly ($p > 0.05$) by Tukey's test. Significance of the *F*-test: Ingredient (I), $p < 0.0001$; Species (S), $p = 0.1101$; I × S, $p = 0.0001$. Coefficient of variation = 1.39%. Digestible protein data were not subjected to statistical analysis.

Blood meal and feather meal had the highest crude protein contents (84.44% and 80.48%, respectively) among all feed ingredients but showed the lowest ADCs for crude protein (45.06% and 66.04%, respectively). Fish meal exhibited the highest protein ADC, likely due to its well-balanced profile of essential amino acids. Its amino acid composition reduces competition for transporters among molecules with similar structures, making it a feed ingredient of high biological value (FAO, 2018; NASCIMENTO T. M. T. *et al.*, 2020).

For tambaqui, fish meal showed the highest ADC of crude protein (87.20%), followed by meat and bone meal (83.08%) and viscera meal (82.68%), without significant differences between the latter two. The lowest values were observed for feather meal (65.21%) and blood meal (46.72%). For tilapia, fish meal (86.38%), viscera meal (85.92%), and meat and bone meal (85.43%) were similar in protein ADC and performed better than feather meal (66.87%) and blood meal (43.39%), which differed significantly.

Although a positive correlation was expected between crude protein content and energy and protein ADCs (SENA, 2012), the low ADC values observed for blood meal and feather meal may be related to the composition and quality of their crude proteins. These ingredients contain amino acid bonds that result in low solubility and, consequently, reduced digestibility (PEZZATO *et al.*, 2012).

Abimorad and Carneiro (2004) reported a crude protein ADC of 57.72% for blood meal in pacu. Furuya *et al.* (2010) found a crude protein ADC of 50.69% for the same ingredient in tilapia, suggesting that tilapia may have a reduced capacity to utilize this protein source compared with pacu. As previously noted, differences in these values for the same feed

ingredient among species are likely related to intrinsic biological characteristics or variations in experimental protocols.

As observed for crude protein digestibility, the ADC of gross energy was also influenced by the interaction ($p < 0.05$) of feed ingredient and species, indicating that gross energy utilization may vary among fish species (Table 3). In practical terms, the efficiency of energy utilization can differ between species, depending on factors such as fish age, feeding habits, and other physiological characteristics (NRC, 2011). The ADCs of gross energy for feed ingredients did not differ significantly between the two species, with the exception of meat and bone meal (82.77% and 89.69% for tambaqui and tilapia, respectively). The mean values observed were 84.91%, 76.17%, 74.73%, and 85.31% for fish meal, blood meal, feather meal, and viscera meal, respectively.

For tilapia, meat and bone meal (89.69%) showed the highest gross energy ADC, followed by fish meal (84.80%) and viscera meal (84.58%), which did not differ from each other. Blood meal (76.70%) and feather meal (73.94%) also did not differ significantly from each other. For tambaqui, the highest coefficients were observed for viscera meal (86.04%), fish meal (85.01%), and meat and bone meal (82.77%), without significant differences among them, followed by blood meal (75.63%) and feather meal (75.52%), which also did not differ from each other (Table 4).

Table 4 – Apparent digestibility coefficients of gross energy and digestible energy contents of animal meal ingredients in diets for tambaqui and Nile tilapia

Species	Meat and bone meal	Fish meal	Blood meal	Feather meal	Viscera meal
Apparent digestibility coefficient (%)					
Tambaqui	82.77 ± 1.47 ^{Ba}	85.01 ± 1.56 ^{Aa}	75.63 ± 1.02 ^{Ab}	75.52 ± 1.86 ^{Ab}	86.04 ± 1.26 ^{Aa}
Nile tilapia	89.69 ± 1.41 ^{Aa}	84.80 ± 1.51 ^{Ab}	76.70 ± 0.60 ^{Ac}	73.94 ± 0.76 ^{Ac}	84.58 ± 1.78 ^{Ab}
Digestible energy content (kcal kg⁻¹)					
Tambaqui	3.141.0 ± 55.9	3.710.5 ± 68.1	4.034.3 ± 54.4	4.076.7 ± 100.3	4.129.9 ± 60.6
Nile tilapia	3.403.6 ± 53.5	3.701.5 ± 66.1	4.091.2 ± 32.2	3.991.0 ± 41.1	4.059.8 ± 85.3

Source: prepared by the authors (2023).

Results are presented as mean ± standard deviation. Means followed by the same uppercase letters within columns or the same lowercase letters within rows do not differ significantly ($p > 0.05$) by Tukey's test. Significance of the *F*-test: Ingredient (I), $p < 0.0001$; Species (S), $p = 0.0752$; I × S, $p = 0.0002$. Coefficient of variation = 1.70%. Digestible energy data were not subjected to statistical analysis.

Overall, the lack of differences in gross energy ADC between the two species, except for meat and bone meal, suggests that a single value of gross energy ADC could be used for each ingredient, regardless of species.

The ADCs of crude protein and gross energy for the evaluated feed ingredients are within the ranges reported in the literature. However, it should be noted that literature values show considerable variation, particularly for feather meal, with crude protein digestibility ranging from 29.12% (PEZZATO *et al.*, 2002) to 78.88% (XAVIER, 2015). For viscera meal, literature values of gross energy ADC range from 69.99% (ABIMORAD; CARNEIRO, 2004) to 89.79% (SENA, 2012).

The low ADCs of crude protein and gross energy observed for blood and feather meals, compared with the other ingredients, may be attributed to their high proportion of low-quality proteins. The digestibility of blood meal (84.44%) and feather meal (80.48%) may be reduced due to their limited solubility caused by disulfide bridges and the presence of sulfur-

containing amino acids. These amino acid bonds, including hydrogen and disulfide bonds, are difficult to break, hindering the action of digestive enzymes. Xavier (2015), in a study investigating the digestibility of feather meal for tilapia, argued that feather meal is primarily composed of keratin, which is highly resistant to proteolytic enzymes and contains large amounts of cystine. The combination of hydrogen bonds and disulfide bridges in cystine contributes to the low digestibility of this protein source.

Feather and blood meals can reduce feed intake when included at high levels in fish diets due to their low palatability. Therefore, their inclusion levels are typically limited to 5%–10%, depending on fish age (FRACALOSI; CYRINO, 2013). According to Brazilian tables for tilapia nutrition, the maximum inclusion levels are 10% for blood meal (FURUYA *et al.*, 2010) and 8% for feather meal (FINKLER *et al.*, 2013). Rocha and Silva (2004) noted that feather meal is considered a low-quality ingredient compared with fish meal because its high contents of cystine and hydrogen bonds limit the action of digestive enzymes. Consistent with this observation and the results of the current study, Narváez-Solarte (2006) reported a protein ADC of 44.07% for different types of blood meal.

It is noteworthy that the inclusion levels of animal meals adopted in the present study were based on experimental protocols used in similar studies (GONÇALVES; CARNEIRO, 2003; MEURER; HAYASHI; BOSCOLO, 2003; ABIMORAD; CARNEIRO, 2004; NARVÁEZ-SOLARTE, 2006). Therefore, it is necessary to extrapolate values to determine minimum digestibility coefficients for the analyzed species and thus avoid potential overestimations.

For the ADCs of total phosphorus, a significant interaction ($p < 0.05$) was observed between feed ingredient and species, indicating that phosphorus utilization may differ between the two fish species (Table 5). Tambaqui showed the highest phosphorus digestibility in blood meal (91.28%), followed by dicalcium phosphate (76.07%), fish meal (68.42%), meat and bone meal (57.13%), and viscera meal (59.72%), without significant differences between the last two ingredients. The lowest value was observed for feather meal (55.71%), which did not differ from meat and bone meal.

Table 5 – Apparent digestibility coefficients of total phosphorus and digestible phosphorus contents of animal meal ingredients in diets for tambaqui and Nile tilapia

Species	Meat and bone meal	Fish meal	Blood meal	Feather meal	Viscera meal
Apparent digestibility coefficient (%)					
Tambaqui	57.13 ± 0.68 ^{Bde}	68.42 ± 1.00 ^{Ac}	91.28 ± 1.43 ^{Aa}	55.71 ± 1.07 ^{Ae}	59.72 ± 0.57 ^{Ad}
Nile tilapia	60.31 ± 0.10 ^{Ad}	68.02 ± 0.99 ^{Ac}	89.44 ± 1.20 ^{Aa}	55.66 ± 0.63 ^{Ae}	60.31 ± 1.80 ^{Ad}
Digestible phosphorus content (%)					
Tambaqui	3.14 ± 0.04	2.25 ± 0.33	0.11 ± 0.00	0.24 ± 0.00	1.61 ± 0.01
Nile tilapia	3.31 ± 0.05	2.24 ± 0.33	0.11 ± 0.00	0.24 ± 0.00	1.63 ± 0.03

Source: prepared by the authors (2023).

Results are presented as mean ± standard deviation. Means followed by the same uppercase letters within columns or the same lowercase letters within rows do not differ significantly ($p > 0.05$) by Tukey's test. Significance of the F-test: Ingredient (I), $p < 0.0001$; Species (S), $p = 0.4276$; I × S, $p = 0.0053$. Coefficient of variation = 1.56%. Digestible phosphorus data were not subjected to statistical analysis.

Similar to tambaqui, tilapia showed the highest ADC of total phosphorus in blood meal (89.44%) and the lowest in feather meal (55.66%). The remaining feed ingredients showed intermediate values of phosphorus ADC.

The data from this study revealed mean digestible protein and energy values of 55.84% and 3705.99 kcal kg⁻¹, respectively, for fish meal, which are higher than those reported in Brazilian tables for tilapia nutrition (46.57% and 3436.13 kcal kg⁻¹, respectively). Digestible phosphorus contents (2.245%) were similar to the table value of 2.37% (FURUYA *et al.*, 2010). These discrepancies may be attributed to a lack of standardization of feed ingredients during processing and differences in the proportions of raw materials in fish diets (GOMES *et al.*, 2007).

Tilapia was able to efficiently digest crude protein in both viscera meal and meat and bone meal. Viscera meal has a similar composition to fish meal (CRUZ-SUÁREZ *et al.*, 2007). Sena (2012) reported that the gross energy ADC in viscera meal for tambaqui was similar to that in fish meal and meat and bone meal. However, the authors found that the ADC of crude protein was highest in fish meal, followed by meat and bone meal and viscera meal, which did not differ from each other. These results corroborate the findings of the current study. Likewise, Guimarães (2006), in analyzing different feed ingredients in tilapia diets, found that the ADC of crude protein in meat and bone meal, fish meal, and viscera meal was 78.44%, 88.60%, and 89.73%, respectively, values closely aligned with those observed in the current study.

Tambaqui possess specialized structures called pyloric caeca. These invaginations of the intestinal wall can be considered an extension of the inner surface of the small intestine. Pyloric caeca allow part of the diet to be retained for a longer period, potentially enhancing the activity of proteolytic enzymes secreted by the pancreas, as well as increasing the area available for nutrient absorption (SEIXAS FILHO *et al.*, 2000; ROTTA, 2003). Based on this anatomy, tambaqui would be expected to utilize feed more efficiently than tilapia. However, this advantage may be offset by the higher intestinal coefficient observed in tilapia (REIS NETO *et al.*, 2018).

Intestinal length can also influence digestibility coefficients, as a longer intestine increases the retention time of feed, allowing digestive enzymes to act more effectively (SOARES *et al.*, 2017; MORAES; ALMEIDA, 2020). Chemical digestion in fish largely depends on the quantity and types of enzymes present in digestive secretions. The distribution and activity of these enzymes may be influenced by numerous factors, such as diet, environment, health status, feeding habits, digestive tract morphology, and species (CORRÊA *et al.*, 2007).

Although both species analyzed here have omnivorous feeding habits, enzyme availability along the digestive tract is expected to vary due to interspecific differences (NRC, 2011). Omnivorous fish can naturally undergo structural and anatomical adaptations in the gastrointestinal tract during development, likely as an adaptation to different feeding habits. This adaptation capacity suggests greater alteration in intestinal length, which can influence the efficiency of nutrient digestion and absorption (BOMFIM; LANNA, 2004; SOARES *et al.*, 2017).

Diet can also influence digestibility coefficients. Silva *et al.* (2022) evaluated the impact of a reference diet on the ADCs of protein ingredients for Nile tilapia. The authors concluded that practical feed ingredients improve crude protein digestibility, particularly those rich in the amino acids arginine, histidine, aspartic acid, and cysteine, which showed higher digestible values. Moreover, the authors noted that amino acid digestibility depends on the

type of protein (high or low biological value) present in feed and the digestive capacity of the animal, depending on the feed's chemical composition.

According to Silva *et al.* (2022), the type of diet (practical, purified, or semi-purified) also affects feed acceptability, with practical diets being more palatable than semi-purified or purified diets. The authors observed that tilapia fed practical diets exhibited higher voracity, which increased voluntary feed intake and, consequently, influenced digestibility results. As emphasized by Moraes and Almeida (2020), increased feed intake enhances substrate availability, stimulating the activity of digestive enzymes and potentially improving nutrient digestibility.

In this study, the reference diet was the same for both species and was formulated with practical ingredients. Therefore, it can be speculated that ADCs obtained herein are close to actual values, as practical diets may stimulate feed intake, leading to greater dilution of endogenous losses. Although feed intake was not measured, the differences observed between species for some ingredients might be attributable to variations in voluntary feed intake (SILVA *et al.*, 2022).

Tambaqui and tilapia occupy a prominent position in the food web, functioning as both primary and secondary consumers. This double role is associated with favorable morphophysiological adaptations that enhance the efficiency of digestion and nutrient absorption for multiple feedstuffs, whether of animal or plant origin (MEURER; HAYASHI; BOSCOLO, 2003; SENA, 2012). These two species also exhibit similar digestive physiology, which may explain the lack of significant differences in the ADCs of crude protein, gross energy, and total phosphorus in most of the tested feed ingredients.

It is essential to maintain a mineral balance in fish diets. Several elements share similar physicochemical properties, which can lead to competition for binding sites on transport molecules. Consequently, an excess of one mineral may impair the absorption of another due to potential saturation of transporters (QUINTERO-PINTO *et al.*, 2011; NRC, 2011; BOMFIM, 2013). Similarly, the absence or limitation of one mineral can affect the absorption and utilization of others (FRACALLOSSI; CYRINO, 2013). Sena (2012) demonstrated that the high mineral content in meat and bone meal negatively affects tambaqui nutrition, showing that the percentage of mineral matter in feed is inversely proportional to its ADC.

It should be noted that, in general, no consistent pattern was observed in variations in digestibility between species. In some cases, tambaqui exhibited higher ADCs, and in others, tilapia. Except for meat and bone meal, digestible contents were similar for both species. Although some feed ingredients differed significantly ($p < 0.05$) between the two species, it is important to consider that the inclusion levels of these feeds are limited, primarily due to their high mineral content (meat and bones, feather and viscera) or low protein digestibility and biological value (blood and feather meals). Therefore, it is recommended to use the mean values for the ADCs of crude protein, crude energy, and total phosphorus for both fish, regardless of statistical differences, since the coefficients of variation obtained in this study were markedly low.

These findings underscore the importance of conducting further studies on nutrition and the nutritional value (digestibility) of feed ingredients used in diets for both fish species. Although tambaqui and Nile tilapia are well-established in national aquaculture, the available

information shows large variability. Furthermore, in the case of tambaqui, information is still scarce. Data on phosphorus digestibility coefficients are particularly limited, making it difficult to formulate feeds with accurate contents to meet the animals' metabolic requirements. The results obtained in this study can serve as a foundation for developing balanced diets with high nutritional quality and greater utilization efficiency, while minimizing the generation of fecal and metabolic waste in the aquatic environment and fish farm wastewaters.

4 FINAL CONSIDERATIONS

In practical terms, and considering the observed exceptions, mean values can be adopted for the digestibility coefficients of animal meal ingredients for both species, as differences were only observed in the ADCs of crude protein in blood meal and viscera meal. Tambaqui showed higher ADCs of crude protein in fish meal and lower ADCs in blood meal. Nile tilapia exhibited higher ADCs of crude protein in fish meal, viscera meal, and meat and bone meal, and the lowest values in blood meal.

Both species showed similar ADCs of gross energy, except in meat and bone meal, for which Nile tilapia had higher digestibility. Tambaqui obtained the highest ADCs of gross energy in viscera, fish, and meat and bone meals, and, like Nile tilapia, showed low values for feather and blood meals.

Regarding total phosphorus, Nile tilapia had the highest ADC in meat and bone meal. For both species, blood meal had the highest coefficient, whereas feather meal showed the lowest.

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STATEMENTS

EACH AUTHOR'S CONTRIBUTION

- **Study conception and design:** Marcos Antonio Delmondes Bomfim, Felipe Barbosa Ribeiro, and Geisiane Silva Sousa.
 - **Data curation:** Rafael Silva Marchão, Geisiane Silva Sousa, Marcos Antonio Delmondes Bomfim, and Daphinne Cardoso Nagib do Nascimento.
 - **Formal analysis:** Marcos Antonio Delmondes Bomfim, Rafael Silva Marchão, Geisiane Silva Sousa, and Jefferson Costa de Siqueira.
 - **Acquisition of financing:** Marcos Antonio Delmondes Bomfim.
 - **Research:** Geisiane Silva Sousa and Rafael Silva Marchão.
 - **Methodology:** Marcos Antonio Delmondes Bomfim, Geisiane Silva Sousa, and Rafael Silva Marchão.
 - **Writing – Initial draft:** Geisiane Silva Sousa.
 - **Writing – Critical review:** Marcos Antonio Delmondes Bomfim and Daphinne Cardoso Nagib do Nascimento.
 - **Proofreading and final editing:** Rafael Silva Marchão.
 - **Supervision:** Marcos Antonio Delmondes Bomfim.
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DECLARATION OF CONFLICTS OF INTEREST

We, **Geisiane Silva Sousa, Marcos Antonio Delmondes Bomfim, Daphinne Cardoso Nagib do Nascimento, Rafael Silva Marchão, Felipe Barbosa Ribeiro, and Jefferson Costa de Siqueira**, declare the following conflicts of interests regarding the manuscript titled "**Digestibility of animal ingredients for tambaqui and Nile tilapia**":

1. **Financial ties:** This work was funded by the Maranhão State Foundation for the Support of Research and Scientific and Technological Development (*Fundação de Amparo à Pesquisa e ao Desenvolvimento Científico e Tecnológico do Maranhão, FAPEMA*).
2. **Professional relationships:** Marcos Antonio Delmondes Bomfim, Felipe Barbosa Ribeiro, Daphinne Cardoso Nagib do Nascimento, and Jefferson Costa de Siqueira work at the Federal University of Maranhão (UFMA), as part of the faculty of the Graduate Program in Animal Science (PPGCA), where Geisiane Silva Sousa was advised by Marcos Antonio Delmondes Bomfim and co-advised by Daphinne Cardoso Nagib do Nascimento during her master's studies. Rafael Silva Marchão is a doctoral student at the Federal University of the São Francisco Valley (UNIVASF) and an external collaborator in graduate research.
3. **Personal conflicts:** Marcos Antonio Delmondes Bomfim and Felipe Barbosa Ribeiro formed a research group focused on topics related to animal nutrition.