

## Determination of Nutrient Composition and Metabolizable Energy for Geese of Paper Mulberry Leaf Powder and Branch-Leaf Powder from Different Origins: Postprint

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### Abstract

This experiment aimed to determine the nutritional composition and goose metabolic energy of paper mulberry leaf powder and paper mulberry branch-leaf powder from different origins. The experiment collected 6 paper mulberry leaf powder samples and 3 paper mulberry branch-leaf powder samples, and their nutritional composition contents were determined using chemical analysis methods. Forty-eight healthy adult male Magang geese at 18 weeks of age with similar body weight were selected and randomly divided into 6 groups, with 8 replicates per group and 1 goose per replicate. Metabolic trials were used to determine the goose metabolic energy of the 6 paper mulberry leaf powder samples and 3 paper mulberry branch-leaf powder samples. The results showed: 1) The average gross energy of the 6 paper mulberry leaf powder samples was 17.07 MJ/kg, and the average contents of crude protein, crude fat, crude ash, crude fiber, calcium, and phosphorus were 22.49%, 2.19%, 17.81%, 13.89%, 3.12%, and 0.54%, respectively; except for gross energy and crude ash content, the coefficient of variation for other nutritional components was greater than 10%. The average gross energy of the 3 paper mulberry branch-leaf powder samples was 18.35 MJ/kg, and the average contents of crude protein, crude fat, crude ash, crude fiber, calcium, and phosphorus were 16.99%, 1.44%, 13.77%, 25.12%, 1.58%, and 0.37%, respectively; crude fat, phosphorus, and crude fiber contents showed large variation, with coefficients of variation greater than 10%. 2) The average contents of tannin, total flavonoids, and water-soluble sugars in paper mulberry branch-leaf powder from different origins were 229.03 g/g, 578.88 g/g, and 4.65%, respectively, with coefficients of variation greater than 20% for total flavonoids and water-soluble sugars. The Hubei paper mulberry branch-leaf powder sample had the highest water-soluble sugar content, while the Anhui pa-

per mulberry branch-leaf powder sample had the highest total flavonoid content. 3) The average apparent metabolizable energy (AME) and true metabolizable energy (TME) of the 6 paper mulberry leaf powders for geese were 9.72 and 10.23 MJ/kg, respectively. The average AME and TME of the 3 paper mulberry branch-leaf powders for geese were 6.87 and 7.35 MJ/kg, respectively. In conclusion, paper mulberry branch-leaf powder has relatively high protein content and nutritional value, making it a good roughage raw material, but the energy utilization efficiency for geese is not high.

## Full Text

### Determination of Nutrient Composition and Metabolizable Energy for Geese of *Broussonetia papyrifera* Leaf Powder and Twig-Leaf Powder from Different Regions

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## Abstract

This study aimed to determine the nutrient composition and metabolizable energy for geese of *Broussonetia papyrifera* leaf powder (BPL) and twig-leaf powder (BPTL) from different regions. Six BPL samples and three BPTL samples were collected and analyzed for nutrient content using chemical methods. Forty-eight healthy 18-week-old male Magang geese with similar body weight were randomly divided into six groups (five test diet groups and one fasting control group), with eight replicates per group and one goose per replicate. The metabolizable energy of the six BPL and three BPTL samples for geese was determined using metabolic trials. The results showed: (1) The average gross energy (GE) of the six BPL samples was 17.07 MJ/kg, and the average contents of crude protein (CP), ether extract (EE), crude ash, crude fiber (CF), calcium (Ca), and phosphorus (P) were 22.49%, 2.19%, 17.81%, 13.89%, 3.12%, and 0.54%, respectively. Except for GE and crude ash, the coefficient of variation (CV) for other nutrient contents exceeded 10%. The average GE of the three BPTL samples was 18.35 MJ/kg, and the average contents of CP, EE, crude ash, CF, Ca, and P were 16.99%, 1.44%, 13.77%, 25.12%, 1.58%, and 0.37%, respectively. The contents of EE, P, and CF showed large variation, with CVs exceeding 10%. (2) The average contents of tannins, total flavonoids, and water-soluble sugars in BPTL from different regions were 229.03 g/g, 578.88 g/g, and 4.65%, respectively, with CVs for total flavonoids and water-soluble sugars exceeding 20%. The Hubei BPTL sample had the highest water-soluble sugar content, while the Anhui sample had the highest total flavonoid content. (3) The average apparent metabolizable energy (AME) and true metabolizable energy (TME) of

the six BPL samples for geese were 9.72 and 10.23 MJ/kg, respectively. The average AME and TME of the three BPTL samples for geese were 6.87 and 7.35 MJ/kg, respectively. In conclusion, BPTL has relatively high protein content and nutritional value, making it a promising roughage material, but its energy utilization by geese is relatively low.

**Keywords:** *Broussonetia papyrifera* leaf powder; *Broussonetia papyrifera* twig-leaf powder; geese; nutrient composition; metabolizable energy

## Introduction

In recent years, China's goose industry has developed rapidly, with feed consumption increasing annually. Insufficient production of feed crops has become one of the factors limiting further industry development, making the importance of developing new feed ingredients increasingly prominent. *Broussonetia papyrifera* is widely distributed in China's Yellow River, Yangtze River, and Pearl River basins, with advantages such as fast growth and strong adaptability, capable of growing in plains, hills, and mountains. As a tree species with extremely strong environmental adaptability and wide-ranging uses, *Broussonetia papyrifera* has high economic and ecological value. If it can be developed as a new feed ingredient, it could both increase the added value of *Broussonetia papyrifera* and reduce feed costs, promoting industry development. In recent years, China's *Broussonetia papyrifera* industry has grown steadily, with national planting area exceeding 1 million mu (1 mu = 666.67 m<sup>2</sup>) and abundant production. *Broussonetia papyrifera* leaves have high crude protein (CP) content, three times that of rice and corn and twice that of wheat, and are rich in various amino acids [1], making them a feed ingredient with high CP content. He Guoying [2] reported that the apparent digestible energy of *Broussonetia papyrifera* leaves for pigs was 10.54 MJ/kg, while for yellow-feathered broilers, the apparent metabolizable energy and true metabolizable energy were 6.92 and 8.11 MJ/kg, respectively. However, domestic research on its application in geese is currently lacking. Therefore, this study collected six BPL samples and three BPTL samples to determine their conventional nutrient composition and metabolizable energy for geese, investigating whether they can serve as quality new feed resources and providing basic data for broader utilization of *Broussonetia papyrifera* resources.

## Materials and Methods

### 1.1 Test Material Preparation

*Broussonetia papyrifera* leaf or leaf powder samples were collected from six regions (Hunan, Hubei, Henan, Hebei, Anhui, and Sichuan) during the same period, with 2 kg of air-dried or sun-dried samples collected from each region, all leaf powder samples collected in autumn. Additionally, three types of BPTL were collected within Guangdong Province, with 2 kg per sample, where Guangdong 1 was collected in autumn, Guangdong 2 in summer, and Guangdong 3

in winter. The collected six BPL and three BPTL samples were crushed and mixed uniformly using the quartering method. The samples were then mixed with corn starch at a certain ratio, with appropriate amounts of multivitamins and minerals and distilled water added before pelleting. After air-drying, the test diets were prepared, sealed in ziplock bags, and stored at  $-4^{\circ}\text{C}$  for later use. A corn starch group was also established as a control.

## 1.2 Experimental Animal Grouping

Forty-eight healthy 18-week-old male Magang geese with similar body weight were randomly divided into six groups (five test diet groups and one fasting control group), with eight replicates per group and one goose per replicate. The metabolic trial was conducted in two batches (nine test diet groups total, one corn starch group, and two fasting control groups), with a parallel fasting control for each batch.

## 1.3 Experimental Animal Management

Before the formal metabolic trial, the selected Magang geese were transferred from outdoor areas to specific goose metabolic cages in the metabolic chamber for a two-week adaptation period. During adaptation, the health and feeding/drinking conditions of the geese were monitored. In the first week of adaptation, feathers around the cloaca were plucked, and a laboratory-made plastic bottle cap (5.5 cm diameter with hollowed-out cover and eight small holes drilled around) was sutured at the cloaca. During adaptation, the geese were fed a formulated diet twice daily (08:00 and 17:00) with free access to water. To ensure consistent experimental conditions, both the pre-test adaptation and formal test periods employed a 14 h (light intensity 20 lx) + 10 h (light intensity 5 lx) lighting regimen. The metabolic chamber was cleaned every three days during adaptation and recovery periods, and thoroughly cleaned 12 h before force-feeding to check whether the sutured caps on the experimental geese were secure.

## 1.4 Metabolic Trial Method

Based on the true metabolizable energy (TME) method, the emptying-force-feeding method was employed using a 24 h + 24 h pattern. According to the digestive physiological characteristics of geese, the specific metabolic trial process was as follows: a two-week adaptation period feeding formulated diets with nipple drinkers for free feed and water intake; a three-day pre-test period feeding test diets; 24 h of fasting and emptying; on the force-feeding day, each goose was force-fed 80 g; excreta were collected accurately for 24 h post-feeding (collected once every 4 h); the fasting group served as a parallel control. Geese had free access to water throughout the metabolic trial.

### 1.5 Excreta Collection and Processing

After force-feeding, excreta were collected in porcelain trays with appropriate amounts of 10% hydrochloric acid added to fix ammonia and prevent volatilization. Samples were dried at 65°C for 58 h, rehumidified for 24 h, weighed and recorded, then crushed and passed through a 40-mesh sieve before being transferred to small ziplock bags and stored at -4°C.

### 1.6 Measurement Indicators and Methods

Conventional nutrient composition was determined for the six BPL samples, three BPTL samples, and excreta. Determination methods for moisture, crude protein, ether extract, calcium, and phosphorus content followed procedures in *Feed Analysis and Feed Quality Detection Technology* (3rd edition). Crude fiber, neutral detergent fiber (NDF), and acid detergent fiber (ADF) content were determined using the filter bag method with an ANKON A200i semi-automatic fiber analyzer according to *Feed Analysis and Feed Quality Detection Technology* (3rd edition). Gross energy was measured using a German IKA C200 automatic calorimeter. Tannin content was determined by acetone extraction-spectrophotometry.

### 1.7 Calculation Formulas

Apparent metabolizable energy (AME, MJ/kg) = [(GE intake - GE excreta) / DM intake] × 10<sup>3</sup>

True metabolizable energy (TME, MJ/kg) = [(GE intake - GE excreta + endogenous GE) / DM intake] × 10<sup>3</sup>

Nutrient apparent utilization (%) = [(total nutrient intake - total nutrient output) / total nutrient intake] × 100

Nutrient true utilization (%) = [(total nutrient intake - total nutrient output + endogenous nutrient output) / total nutrient intake] × 100

### 1.8 Data Processing

Data were organized and preliminarily processed using Excel 2007, then subjected to one-way ANOVA using SPSS 20.0 software, followed by Duncan's multiple comparison test and independent samples t-test. The statistical significance level was set at P < 0.05, and experimental results were expressed as "mean ± standard error."

## Results

### 2.1 Nutrient Composition of BPL and BPTL

As shown in Table 1, the gross energy of the six BPL samples ranged from 16.28 to 18.13 MJ/kg, with an average of 17.07 MJ/kg; crude protein content ranged from 17.78% to 26.47%, averaging 22.49%; crude fiber content ranged from 11.89% to 16.35%, averaging 13.89%; and the average contents of ether

extract, crude ash, calcium, and phosphorus were 2.19%, 17.81%, 3.12%, and 0.54%, respectively. Except for gross energy and crude ash, the coefficient of variation for other nutrient contents exceeded 10%. For the three BPTL samples, gross energy ranged from 17.94 to 18.76 MJ/kg, averaging 18.35 MJ/kg; crude protein content ranged from 15.80% to 17.89%, averaging 16.99%; crude fiber content ranged from 21.25% to 27.24%, averaging 25.12%; and the average contents of ether extract, crude ash, calcium, and phosphorus were 1.44%, 13.77%, 1.58%, and 0.37%, respectively. Ether extract, phosphorus, and crude fiber contents showed large variation, with coefficients of variation exceeding 10%. These results indicate that compared with BPL, BPTL has higher gross energy and crude fiber content but lower crude protein, ether extract, calcium, and phosphorus contents.

## 2.2 Bioactive Substance Content in BPL and BPTL

As shown in Table 2, water-soluble polysaccharide content in BPL from different regions varied significantly, with an average of 5.11% and the highest content of 8.14% in the Hubei sample. Total flavonoid content also varied significantly, averaging 541.89 g/g with the highest content of 890.29 g/g in the Anhui sample. Tannin content in BPL averaged 233.07 g/g. In BPTL, water-soluble polysaccharide content averaged 3.73%, total flavonoid content averaged 652.89 g/g, and tannin content averaged 216.91 g/g. These findings demonstrate that BPTL has higher average total flavonoid content than BPL, but lower average water-soluble polysaccharide and tannin contents.

## 2.3 Metabolizable Energy and Energy Utilization of BPL and BPTL for Geese

As shown in Table 3, the apparent metabolizable energy (AME) of the six BPL samples for geese ranged from 7.49 to 10.94 MJ/kg, averaging 9.72 MJ/kg; true metabolizable energy (TME) ranged from 8.21 to 11.47 MJ/kg, averaging 10.23 MJ/kg; apparent energy utilization ranged from 43.89% to 67.21%, averaging 57.34%; and true energy utilization ranged from 48.10% to 70.46%, averaging 60.31%. For the three BPTL samples, AME ranged from 5.12 to 8.04 MJ/kg, averaging 6.87 MJ/kg; TME ranged from 5.17 to 8.58 MJ/kg, averaging 7.35 MJ/kg; apparent energy utilization ranged from 27.28% to 44.84%, averaging 37.60%; and true energy utilization ranged from 27.58% to 47.85%, averaging 40.24%. These results clearly show that BPL has higher metabolizable energy and energy utilization than BPTL.

## Discussion

### 3.1 Nutrient Composition of BPL and BPTL

Various domestic and international scholars have studied the nutritional value of *Broussonetia papyrifera*, though its nutritional value is significantly affected by variety, growing environment, and season. Tu Yan et al. [3] determined

that *Broussonetia papyrifera* leaves contained 26.05% crude protein and 3.35% calcium, along with trace elements such as iron, manganese, and zinc, making them a quality feed ingredient. Obour et al. [4] reported that *Broussonetia papyrifera* leaves contained 27.17% crude protein, 6.45% ether extract, 2.54% calcium, and 0.23% phosphorus on a dry matter basis, along with potassium, magnesium, and other trace elements. Yang Zuda et al. [5] collected *Broussonetia papyrifera* leaves and determined their moisture content as 13.0%, crude protein as 24.0%, ether extract as 3.0%, crude fiber as 11.7%, and calcium and phosphorus contents as 2.7% and 0.3%, respectively. Compared with conventional feed ingredients such as rice and corn, its crude protein content is higher than rice, corn, and wheat, second only to soybean; its ether extract content is equivalent to corn, twice that of rice and wheat, but lower than soybean, indicating high nutritional value. Xu Youxin et al. [6] collected *Broussonetia papyrifera* leaves from the same location in different seasons (spring, summer, autumn) and determined their nutrient contents, finding crude protein content of 18.4%–25.3%, ether extract of 5.9%–6.9%, and crude fiber of 12.6%–18.0%, with ether extract and crude fiber increasing and crude protein decreasing with seasonal changes. Yu Ming et al. [7] determined that *Broussonetia papyrifera* leaves collected in July, August, and September showed highest crude protein in July and highest crude fiber, calcium, and phosphorus in September. They also found that leaves from the upper one-third of branches had higher crude protein, while leaves from the lower one-third had higher crude fiber, calcium, and phosphorus. As leaf age increased, more calcium and phosphorus were deposited, resulting in higher contents, while crude fiber increased and crude protein relatively decreased. Due to differences in collection locations and seasons among samples in this study, nutrient contents varied accordingly. Guangdong 2 sample, collected in summer, had higher crude protein than Guangdong 3 sample collected in winter, consistent with literature trends. *Broussonetia papyrifera* leaf samples had high calcium and low phosphorus contents, requiring attention to dietary calcium-phosphorus ratios in practical applications. Both BPL and BPTL had crude ash contents above 10%, indicating high mineral content, which is consistent with literature reporting that *Broussonetia papyrifera* leaves are rich in various mineral elements. The crude protein, ether extract, crude fiber, and calcium contents of BPL samples in this study were consistent with previously reported results. While BPL had higher nutrient content than BPTL, BPL is relatively more difficult to collect than BPTL, making BPTL more suitable for large-scale development and utilization.

*Broussonetia papyrifera* is rich in flavonoids, with more than 40 types isolated from different plant parts [8]. Yang Xueying [9] conducted qualitative analysis and total content determination of components in *Broussonetia papyrifera* leaf extracts, finding flavonoids as the main components with certain antioxidant properties that enhanced with increasing concentration. Xiong Yanfei et al. [10] demonstrated that flavonoids have strong scavenging capacity for hydroxyl radicals and superoxide anions. Li Yong [11] found that adding 5% *Broussonetia papyrifera* leaves to diets significantly reduced plasma total chole-

terol (TC), glucose (GLU), triglycerides (TG), and free fatty acids (NEFA) in obese mice, while adding 2% and 5% leaves significantly increased plasma high-density lipoprotein (HDL) content, accelerating HDL transport of peripheral cholesterol and promoting liver conversion and excretion of cholesterol. Cui Can et al. [12] found that *Broussonetia papyrifera* leaf extracts had inhibitory effects on dermatophytic fungi such as *Trichophyton rubrum*, *Candida krusei*, *Fonsecaea compacta*, and *Trichophyton mentagrophytes*, with obvious and stable effects.

The main anti-nutritional factor in *Broussonetia papyrifera* leaves is tannin, a class of water-soluble phenolic compounds that can form complexes with carbohydrates [13], proteins [14], and metal ions [15] in the animal digestive tract, resulting in compounds that are difficult to absorb and leading to reduced feed utilization and impaired animal growth. Yu Ming et al. [7] determined tannin contents in *Broussonetia papyrifera* leaves collected in July, August, and September as 0.95%, 1.34%, and 2.36%, respectively. Obour et al. [4] reported tannin contents of 6.96%–7.09% in *Broussonetia papyrifera* leaves. The relatively low tannin content measured in this study may be related to the determination method. Traditionally, physicochemical or biodegradation methods have been used to reduce tannin content in plants. Studies have shown that treating two types of tree leaves with different concentrations of sodium hydroxide (NaOH) solution for 7 days resulted in linear decreases in tannin content with increasing NaOH concentration [16]. Fungal fermentation can significantly reduce tannin content in black locust while increasing crude protein content and significantly improving dry matter digestibility [17]. However, recent studies have shown that adding appropriate amounts of tannins to diets does not affect animal growth and may improve antioxidant capacity [18–20], inhibit parasites [21,22], and suppress harmful bacteria [23], benefiting animal growth.

### 3.2 Metabolizable Energy and Energy Utilization of BPL and BPTL for Geese

China has a long history of using woody plants as animal feed, and increasing attention has been paid to the prospects of woody plants as new feed ingredients, with numerous related studies. Wang Yongchang et al. [24] reported that the TME of mulberry twig-leaf powder for Magang geese was 4.92 MJ/kg, with true energy utilization of 26.91%. Wang Zenghuang et al. [25] found that the AME and TME of banana stem-leaf powder for Magang geese were 6.50 and 7.20 MJ/kg, respectively, with apparent energy utilization of 37.50% and true energy utilization of 41.15%. Geese have low energy utilization for these high-fiber, low-protein woody plants. This study determined that the AME of BPTL for geese ranged from 5.12 to 8.04 MJ/kg, TME from 5.17 to 8.58 MJ/kg, apparent energy utilization from 27.28% to 44.84%, and true energy utilization from 27.58% to 47.85%. The low energy utilization for high-fiber, low-protein BPTL observed in this study is consistent with related reports.

Compared with BPTL, geese have higher energy utilization for BPL. This study

determined that the AME of BPL from different regions for Magang geese ranged from 7.49 to 10.94 MJ/kg, TME from 8.21 to 11.40 MJ/kg, apparent energy utilization from 43.89% to 67.21%, and true energy utilization from 48.10% to 70.46%. He Guoying [2] reported that the digestible energy of *Broussonetia papyrifera* leaves for pigs was 10.54 MJ/kg. For three-yellow chickens, the AME was 6.92 MJ/kg, TME was 8.11 MJ/kg, and true energy utilization was 50.71%. Geese have higher energy utilization for *Broussonetia papyrifera* leaf powder than chickens. Wang Ruixiao et al. [26] demonstrated that geese have higher energy utilization than chickens for high-fiber feed ingredients. Currently, there are few domestic and international reports on the energy utilization of *Broussonetia papyrifera* leaves in animals, and metabolic studies in geese are particularly lacking, necessitating further investigation into the reasons for relatively low energy utilization.

## Conclusion

1. *Broussonetia papyrifera* leaf powder has higher nutritional value than twig-leaf powder, with crude protein content reaching up to 26.47%, making it a quality protein supplement feed ingredient.
2. Geese have higher energy utilization for *Broussonetia papyrifera* leaf powder than for twig-leaf powder. The average AME of BPL for geese was 9.72 MJ/kg, with a maximum of 10.94 MJ/kg. The average AME of BPTL for geese was 6.87 MJ/kg, with energy utilization below 50%, indicating relatively low energy utilization.

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