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Productive and metabolic increments of the inclusion of *Broussonetia papyrifera* to replace maize silage in growing goats

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Abstract: The study was conducted to investigate the effects of *Broussonetia papyrifera* (BP) on growth performance, rumen fermentation, digestion and meat quality in Huanghuai white goats. Diets were developed with increasing doses of BP to replace maize silage [0% (control), 15% (BP15), 30% (BP30) and 45% (BP45)]. The design was completely randomized with 4 groups of 10 animals each, totalizing 40 animals (aged 150 ± 10 days and body weight of 25.32 ± 0.52 kg). Final body weight, average daily gain and average daily feed intake increased (linear and quadratic, $P < 0.05$) as BP increased in the diet. The highest body weight and average daily gain were seen in the BP30 group. Feed conversion ratio also improved significantly (linear and quadratic, $P < 0.05$) with the increasing level of BP. Although the concentration of microbial protein (MCP) in a linear ($P = 0.0028$) and quadratic ($P = 0.0035$) fashion with increasing level of BP, however, NH_3-N in a quadratic ($P = 0.0841$) fashion with increasing level of BP. Crude protein was increased with increasing BP in the diets (linear and quadratic, $P < 0.05$). Nitrogen intake, urinary excretion nitrogen and nitrogen retention were significantly improved (linear and quadratic, $P < 0.05$) with dietary BP addition in goats. Dietary BP supplementation increased pH (linear and quadratic, $P < 0.05$) and tended to reduce L* (lightness) (linear and quadratic, $P < 0.05$). Additionally, the redness value of meat was significantly improved by BP addition (linear and quadratic, $P < 0.05$). The inclusion of BP in the goat diet promotes growth and increases the digestibility of crude protein in addition to improving the meat colour and rumen fermentation.

Keywords: goat; nutrient digestibility; meat quality; fermentation; pH

Broussonetia papyrifera (BP) belongs to the genus Broussonetia of the mulberry family, which is widely distributed in China, Japan, India and other countries (Penailillo et al. 2016). *Broussonetia papyrifera* (BP) is characterized by rapid growth, strong adaptability, wide distribution, easy reproduction, high heat tolerance and short rotation period (Adigbli et al. 2019; Qureshi et al. 2020). The leaves of BP contain 21.6% of crude protein, 4.3% of ether extract, and 1.9% of calcium (Wu et al. 2019). They are suitable for feeding cattle, sheep, rabbits and other herbivores, and convenient for large-scale

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intensive production. With the rapid development of animal husbandry, the output of feed resources has not been increased correspondingly. As a new feed resource, the planting area of BP is also growing, which can alleviate this problem to a certain extent (Sun et al. 2012).

Broussonetia papyrifera has recently been used in diets for growth-finishing pigs [17% dry matter (DM)] (Zhang et al. 2015), growing rabbits (20% DM) (Wu et al. 2019) and lambs (10% DM) (Ma et al. 2014). Wu et al. (2019) found that replacing lucerne meal with BP can improve growth performance and meat colour of New Zealand White rabbits. Adding 15% of BP to cattle diet can improve meat quality, growth performance, antioxidant function and meat quality (Si et al. 2018).

Mengmeng et al. (2018) found that BP could replace 30% of silage maize as sheep feed. At present, there are few reports on the application of BP leaves in goat diets, so we hypothesized that diets with BP can improve the quality of meat and the performance of goats. The objective of the present study was to evaluate the effects of replacing silage maize with BP leaves on growth performance, rumen fermentation characteristics, meat quality and nutrient utilization in goats.

MATERIAL AND METHODS

The experiment was conducted on the goats farm of Anhui Lixin Lvshu Animal Husbandry Co., Ltd. (Fuyang, P.R. China, 115°54'E, 32°51'N) in September 2019. The experimental protocol was reviewed and approved by the Animal Care and Use Committee of Anhui University of Science and Technology (Fengyang, P.R. China).

Animals and diets

Forty Huanghuai white goats, aged 150 ± 10 days and body weight of 25.32 ± 0.52 kg, were randomly assigned to four treatments with 10 replicates. The branches of the biennial *Broussonetia* plants were harvested when they reached 60 cm in length. The animals in the control treatment were fed a maize silage-based diet, and the three treatment groups were fed a similar diet in which the silage maize was replaced by 15% (BP15), 30% (BP30), or 45% (BP45) BP. Throughout this study, the four flocks received a ration with a forage to concentrate ratio of 70 : 30. The ingredients and the chemical composition of experimental diets are given in Table 1.

The temperature was 17 ± 7 °C and the relative humidity was 60 ± 15%. The goats had free access to clean drinking water. The experimental animals were kept individually in 1.0 m × 1.5 m wooden pens with steel floors. After a 60-day adaptation period, goats were housed with different experimental diets, with total collection of faeces and urine during the final seven days of the experiment.

Experimental protocol and sample collection

All goats were weighed at the beginning and end of the feeding trial. Feed provided and rejected was recorded daily throughout the entire experimental period. The average daily gain, dry matter intake and feed conversion rate were calculated for each experimental unit.

Table 1. Ingredients and chemical composition of experimental diets

Item	Control	BF15	BF30	BF45
Ingredient (g/kg)				
Corn silage	300	255	210	165
BP	–	45	90	135
Corn	440	440	440	440
Bran	70	70	70	70
Soybean meal	140	140	140	140
Dicalcium phosphate	7	7	7	7
Sodium bicarbonate	7	7	7	7
NaCl	6	6	6	6
Mineral-vitamin premix[1]	30	30	30	30
Nutrient content[2] (% DM)				
ME (MJ/kg)	6.53	6.54	6.56	6.59
CP (g/kg)	102.43	103.65	104.91	106.29
Ca (g/kg)	7.39	7.78	8.21	8.96
P (g/kg)	3.32	3.42	3.51	3.63

BP = *Broussonetia papyrifera*; CP = crude protein; DM = dry matter; ME = metabolisable energy

[1]The premix provided per kg of concentrated feed: vitamin E 1 480 IU, vitamin A 60 000 IU, vitamin D3 81 000 IU, $CuSO_4$ 90 mg, $FeSO_4$ 354.18 mg, $ZnSO_4$ 274.5 mg, K_2SO_4 255.8 mg, $CoCl_2$ 1.25 mg, $MnSO_4$ 230.13 mg, $NaHCO_3$ 3 704.55 mg, Na_2SeO_3 45.55 mg; [2]Calculated based on analysed nutrients of each ingredient

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During the final seven days of the experiment, the daily intake of each goat was measured. During the experimental period, urine was separated from faeces by a separator attached to the bottom of the metabolic cage. Faecal and urine samples were collected and their quantity was measured daily. During the seven days, 5% of feed, 10% of urine and 20% of faeces were collected and frozen at −20 °C to determine the apparent digestibility of nutrients.

At the end of the feeding trial, all goats were euthanized according to the regulations of Animal Protection and Use Committee of Anhui University of Science and Technology. Meanwhile, rumen fluid was sampled after eviscerating and filtered through four layers of cheese cloth for the determination of pH, microbial protein (MCP) and ammonium nitrogen (NH_3-N). The whole *longissimus lumborum* muscle was taken for meat quality analysis.

Chemical analysis

Dry matter (DM) of feed, leaf meals and faeces was measured after drying in the oven at 65 °C for 48 h, grinding, and filtering through a 1 mm screen for chemical analysis. The chemical composition data on DM were corrected for the residual moisture after 3 h at 105 °C. The ash, crude protein and ether extract contents were determined by the methods of the Association of Official Analytical Chemists (AOAC 1995). The organic matter content was calculated as the weight loss upon ashing. The neutral and acid detergent fibres were determined using the method of Van Soest et al. (1991) with an ANKOM A200i fibre analyser (ANKOM Technology, Macedon, USA). The NH_3-N concentration was measured by the method described by Broderick and Kang (1980), and MCP was measured as described by Salem et al. (2015).

Statistical analysis

Data were analysed as a completely randomised design using the General Linear Models procedures of SAS v9.1.3. The following model was used:

$$Y_{ij} = \mu + A_i + e_{ij}$$

where:

Y_{ij} – the observation;

μ – the overall mean;

A_i – the effect of *Broussonetia papyrifera* level;

e_{ij} – the random error.

Duncan's multiple test was used to test the differences between the treatment groups, and significant differences were accepted if $P < 0.05$. Linear and quadratic effects due to BP level were determined.

RESULTS

Growth performance

The growth performance of goats was significantly improved ($P < 0.05$) by dietary BP addition (Table 2). Furthermore, the final body weights and daily gain were higher ($P < 0.05$) in BP30 treatments compared to BP15 and BP45 treatments. In addition, feed conversion as feed intake/weight gain was increased ($P < 0.05$) in BP30 treatments compared to BP15 treatments. Final body weight,

Table 2. Influence of dietary *Broussonetia papyrifera* (BP) on growth performance in goats

Item	Control	BP15	BP30	BP45	Pooled SEM	*P*-value	
						linear	quadratic
Initial body weight (kg)	23.58	23.52	23.58	23.62	0.18	0.812 4	0.938 6
Final body weight (kg)	25.92^{c}	27.54^{b}	29.25^{a}	27.91^{b}	0.35	0.000 4	0.000 1
Average daily gain (g/head per day)	39.15^{c}	67.15^{b}	94.56^{a}	71.44^{b}	5.55	0.000 5	0.001
Average daily feed intake (g/head per day)	735.39^{b}	880.99^{a}	963.88^{a}	907.33^{a}	42.84	0.007 1	0.002 5
Feed conversion ratio[1]	20.21^{a}	14.16^{b}	10.30^{c}	13.02^{bc}	0.92	0.000 1	0.000 1

BP15 = 15% supplemental BP; BP30 = 30% supplemental BP; BP45 = 45% supplemental BP

$^{a-c}$Means within a row with different superscripts differ ($P < 0.05$)

[1]Feed conversion ratio = kg feed ingested/kg of body weight gain

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Table 3. Influence of dietary *Broussonetia papyrifera* (BP) on rumen fermentation characteristics in goats

Item	Control	BP15	BP30	BP45	Pooled SEM	*P*-value	
						linear	quadratic
pH	5.63[c]	5.98[b]	6.21[a]	5.95[b]	0.06	0.002 6	0.000 1
NH_3-N (mg/ml)	4.13	4.45	4.40	4.27	0.10	0.447 9	0.084 1
MCP (mg/ml)	20.21[b]	21.77[ab]	22.83[a]	22.59[a]	0.54	0.002 8	0.003 5

BP15 = 15% supplemental BP; BP30 = 30% supplemental BP; BP45 = 45% supplemental BP; MCP = microbial protein; NH_3-N = ammonium nitrogen

[a–c]Means with different letters within a row differ significantly ($P < 0.05$); $n = 10$ per treatment

average daily gain and average daily feed intake increased (linear and quadratic, $P < 0.05$) as BP increased in the diet, the highest body weight and average daily gain were seen in the BP30 group. The feed conversion ratio was also improved significantly (linear and quadratic, $P < 0.05$) with the increasing level of BP.

Rumen fermentation

The pH in the rumen increased (Table 3, linear and quadratic, $P < 0.05$) as BP increased in the diet; the highest pH was seen in the BP30 group. Although the concentration of MCP in a linear ($P = 0.002\ 8$) and quadratic ($P = 0.003\ 5$) fashion with increasing level of BP, however, NH_3-N in a quadratic ($P = 0.084\ 1$) fashion with increasing level of BP. In addition, the concentration of MCP was higher ($P < 0.05$) for the BP30 and BP45 treatments when compared to the control.

Digestion of nutrients and N utilisation

There was no treatment effect on the digestibility of DM, neutral detergent fibres and acid detergent fibres ($P > 0.10$; Table 4). Crude protein was increased with increasing BP in the diets (linear and quadratic, $P < 0.05$). Nitrogen intake, urinary excretion nitrogen and nitrogen retention were significantly improved (linear and quadratic, $P < 0.05$) with dietary BP addition in goats. Comprehensive analysis showed that BP30 had the best utilization of nitrogen.

Meat quality

Drip loss, b* (yellowness), and shear force were not affected by dietary BP addition ($P > 0.10$). Dietary BP supplementation increased pH (linear and quadratic, $P < 0.05$; Table 5) and tended to reduce L* (linear and quadratic, $P < 0.05$).

Table 4. Influence of dietary *Broussonetia papyrifera* (BP) on digestibility and N utilisation in goats

Item	Control	BP15	BP30	BP45	Pooled SEM	*P*-value	
						linear	quadratic
Digestibility (%)							
Dry matter	73.13	73.71	74.40	74.53	0.61	0.089 7	0.226 9
Crude protein	67.25[b]	68.22[ab]	70.36[a]	70.55[a]	0.77	0.001 7	0.007 0
Acid detergent fibre	79.37	76.35	80.20	76.08	3.55	0.717 2	0.927 3
Neutral detergent fibre	71.72	72.90	74.68	73.26	1.74	0.433 8	0.572 5
Nitrogen utilization (g/day)							
Nitrogen intake	12.48[b]	14.95[a]	16.45[a]	15.76[a]	0.67	0.001 4	0.000 6
Fecal nitrogen	4.07[b]	4.77[ab]	4.87[a]	4.65[ab]	0.24	0.120 2	0.057 6
Urinary nitrogen	3.96[b]	4.19[ab]	4.20[ab]	4.67[a]	0.16	0.007 2	0.022 3
Nitrogen retention	4.45[c]	5.99[b]	7.38[a]	6.45[ab]	0.44	0.001 9	0.000 3

BP15 = 15% supplemental BP; BP30 = 30% supplemental BP; BP45 = 45% supplemental BP

[a–c]Means with different letters within a row differ significantly ($P < 0.05$); $n = 10$ per treatment

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Table 5. Influence of dietary *Broussonetia papyrifera* (BP) on meat quality in goats

Item	Control	BP15	BP30	BP45	Pooled SEM	P-value	
						linear	quadratic
pH	6.03[b]	6.23[a]	6.24[a]	6.32[a]	0.05	0.001 3	0.003 4
L*	39.40[a]	38.92[ab]	37.89[ab]	37.51[b]	0.54	0.010 0	0.037 7
a*	19.46[b]	20.26[a]	21.05[a]	21.00[a]	0.26	0.000 1	0.000 1
b*	6.48	6.60	6.61	6.59	0.06	0.256 4	0.308 9
Drip loss (%)	1.80	1.79	1.82	1.78	0.02	0.778 7	0.806 8
Shear force (kgf)	64.15	65.08	64.34	68.00	2.27	0.309 8	0.510 0

a* = redness; b* = yellowness; BP15 = 15% supplemental BP; BP30 = 30% supplemental BP; BP45 = 45% supplemental BP; L* = lightness

[a,b]Means with different letters within a row differ significantly ($P < 0.05$); $n = 10$ per treatment

Additionally, the redness value of meat was significantly improved by BP addition (linear and quadratic, $P < 0.05$).

DISCUSSION

Growth performance

The results of the present study showed that dry matter intake was significantly higher in goats fed diets based on BP than in goats fed maize silage. Studies have shown that BP is very palatable to sheep and goats (Obour and Oppong 2015). With the increase in the proportion of BP replacing silage maize, the average daily gain of goats increased significantly, and the feed utilization efficiency was significantly improved. Compared to silage maize, BP can provide more balanced amino acids to the goat and has a higher digestibility and utilization rate of high quality crude fibre (Li and Guo 2009; Wu et al. 2019). The results showed that the replacement of silage maize by BP (30%) can promote growth.

Rumen fermentation

The pH value of rumen fluid is an important indicator to reflect the environmental condition of rumen and the fermentation degree of feed in rumen. Rumen pH is constantly changing, and the magnitude of this change is dependent on the type of feed (level of concentrate, physically effective neutral detergent fibres etc.) and feeding time. Ruminal pH has been reported to vary between 5.5 and 7.5 (Seankamsorn et al. 2017). In this experiment, the pH of rumen fluid was between 5.52 and 6.32. Weak acidity provides the best conditions for rumen microorganisms to survive, which is conducive to the digestion and degradation of fibre and production of volatile fatty acids (Kingamkono et al. 1994). Degradation of starch and cellulose by rumen microorganisms to produce volatile fatty acids will reduce the rumen pH, while the pH value of rumen would be increased by degrading protein and producing ammonia (Wang et al. 2016; Asizua et al. 2018). The BP has higher crude protein content than silage, but lower starch content. With the increase of BP, the proportion of silage maize decreased, less ruminal digestible carbohydrates were ingested, so the rumen pH value increased.

Ammonia nitrogen is the end product of rumen feed peptide, amino acid, protein, urea and non-protein nitrogen decomposition, and it is also the main raw material of rumen microbial protein synthesis (Cantalapiedra-Hijar et al. 2018; Patra and Aschenbach 2018). Too high or too low concentration of ammonia nitrogen in rumen is not conducive to the growth of rumen microorganisms. Therefore, maintaining the optimal concentration of ammonia nitrogen in rumen fluid is the primary condition to ensure protein synthesis (Bayat et al. 2017). The most suitable concentration of ammonia nitrogen in rumen fluid is 0.35–29 mg/dl (Owens and Bergen 1983). The concentration of MCP reflects the ability of microorganisms to utilize ammonia nitrogen and the quantity of the microbial population in rumen (Phillipson et al. 1959; Li et al. 2017). Many factors restrict the synthesis of MCP (Valente et al. 2016). The pH value is the key factor to limit MCP synthesis (Voelker and Allen 2003). Rumen digestible protein and carbohydrate are two main factors affecting

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MCP (Arroquy et al. 2004). In addition, the decrease of pH value in rumen can inhibit the growth and reproduction of microorganisms, which leads to the decrease of protozoa number in rumen and the change of microbial population structure (Belenguer et al. 2010). In this study, the concentration of ammonia nitrogen in rumen fluid was 4.03–4.26 mg/dl. The results showed that the concentration of ammonia nitrogen (quadratic, $P = 0.084\,1$) and MCP increased with the increase in the proportion of BP instead of silage maize, which was related to the higher protein content of BP than in silage maize. In addition, the feed intake was significantly improved by dietary BP addition in goats. Lees et al. (1990) showed that the high protein diet could increase the concentration of ruminal ammonia nitrogen.

Digestion of nutrients and N utilisation

Silivong (2012) showed that the paper mulberry has high digestibility of organic matter and crude protein, and it also has a high nutritional value. The high content of crude protein in BP can provide a good nitrogen source for rumen microorganisms (Tu et al. 2009). The high content of fibre in silage maize can provide a carbon source for rumen microorganisms (Ivkovic et al. 2019; Toth et al. 2019). The combination of the two meets the energy to nitrogen ratio required for rumen microorganisms, BP can provide rumen microorganisms with nutrients needed for growth, improve the growth environment of rumen microorganisms, accelerate food decomposition and improve digestibility. Therefore, the energy and nitrogen balance of 30% diet of BP instead of silage maize is better, which shows that the apparent nutrient digestibility of goats in the corresponding experimental group is higher.

Charbonneau et al. (2006) showed that the imbalance of the ratio of carbohydrate to crude protein in the diet will lead to the decrease of nitrogen utilization rate in the process of rumen microbial degradation. The results showed that when the proportion of *Broussonetia papyrifera* in roughage was 30%, the amount of nitrogen deposition and nitrogen utilization rate in the body of Huanghuai Baishan sheep were higher than those in the control group. When the proportion of BP in roughage was 60%, the urinary nitrogen increased and the nitrogen utilization rate decreased, which may be related to the regulation of energy nitrogen balance in animals.

Meat quality

Muscle water-holding capacity is an important factor to measure the quality of meat (den Hertog-Meischke et al. 1997). The lower the drip loss rate and the smaller the shear force, the better the mutton quality (Kim et al. 2015). Meat colour is the most intuitive indicator of meat quality, and it is also an indicator of consumer concern (Powell et al. 1996). The results showed that adding BP to the diet significantly increased the redness of meat, but it had no significant effect on the water-holding capacity of meat. In the normal range, the increase of redness will increase consumers' purchase desire. After slaughtering sheep, haemoglobin was lost greatly, myoglobin became the main reason of red meat (Hosseini et al. 2019). The higher the myoglobin content in muscle, the greater the red degree of meat colour (Li et al. 2018). Wu et al. (2019) found that the iron content of BP was higher than that of maize, and supplementation of BP had a positive effect on the red value of rabbit meat.

Conflict of interest

The authors declare no conflict of interest.

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