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Effect of *Broussonetia papyrifera L.* (paper mulberry) silage on dry matter intake, milk composition, antioxidant capacity and milk fatty acid profile in dairy cows

Bingwen Si[1,2,a], Hui Tao[1,a], Xiaoli Zhang[3], Jiangpeng Guo[4], Kai Cui[1], Yan Tu[1], and Qiyu Diao[1,*]

* **Corresponding Author:** Qiyu Diao
Tel: +1-336-285-4807, **Fax:** +1-336-334-7288,
E-mail: soh@ncat.edu

[1] Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing 10081, China
[2] National Engineering Research Center of Biological Feed, Beijing 10081, China
[3] Beijing Plant Protection Station, Beijing 100029, China
[4] Beijing Animal Husbandry Station, Beijing 100029, China

[a] These authors contributed equally to this research and should be considered co-first authors.

ORCID
Bingwen Si
https://orcid.org/0000-0001-5050-7078
Hui Tao
https://orcid.org/0000-0003-3981-9251
Xiaoli Zhang
https://orcid.org/0000-0002-2969-7337
Jiangpeng Guo
https://orcid.org/0000-0002-0786-064X
Kai Cui
https://orcid.org/0000-0002-6885-952X
Yan Tu
https://orcid.org/0000-0002-4324-6188
Qiyu Diao
https://orcid.org/0000-0002-8037-1471

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Objective: This study was carried out to investigate the possible application of *Broussonetia papyrifera* (*B. papyrifera*) silage as a functional feeding stuff in dairy cattle.
Methods: Seventy-two Holstein cows were divided into four groups randomly and allocated to 6 pens with 3 individuals in each group and fed the original total mixed ratio (TMR) in the dairy farm or the new TMR with 5%, 10%, and 15% *B. papyrifera* silage, separately. Feed intake were recorded, milk and blood samples were collected, and milk composition, blood metabolites and milk fatty acids composition were measure at the end of the experiment.
Results: Dry matter intake of cows decreased when they fed on diet with *B. papyrifera*, but no differences were observed in body condition score, milk yield, milk protein and lactose, feed efficiency and serum metabolites between groups. Both 10% or 15% of *B. papyrifera* silage in the diet significantly increased the immunoglobulin A (IgA) and IgG in serum, 15% of *B. papyrifera* silage increased the content of serum catalase, superoxide dismutase, total antioxidant capacity, and decreased the content of 8-hydroxy-2'-deoxyguanosine. Furthermore, 10% or 15% of *B. papyrifera* silage resulted in a significant decrease in the milk somatic cell count, and increased the polyunsaturated fatty acids content in the milk.
Conclusion: The diets with 10% to 15% of *B. papyrifera* silage might enhance the immune and antioxidant function of dairy cows and increase the polyunstaturated fatty acid concentration in the milk.

Keywords: *Broussonetia papyrifera* Silage; Immune Function; Antioxidant Capacity; Milk; Dairy Cows

INTRODUCTION

Animal husbandry practices are closely linked with animal health, production and welfare. Intensive model of farm animal husbandry brings high yields, but compromises animal health and welfare which might increase the incidence of diseases. For ruminants, there are great opportunities to explore antioxidant enriched forage.

Broussonetia papyrifera L. (*B. papyrifera*, paper mulberry) is a deciduous tree or shrub in the family Moraceae that is native to eastern Asia, and widespread in China, which can be used as a food or a source of traditional medicine to treat various diseases for both humans and animals. Also the *B. papyrifera L.* fruits polysaccharides have antioxidant and antibacterial activities [1]. And it is reported that the radix of *B. papyrifera L.* had the greatest antinociceptive and anti-inflammatory effects when different parts of the plant were compared as treatment for chemical-induced pain and inflammation in rodents [2]. *B. papyrifera L.* had a protective effect against hydrogen peroxide-induced oxidative stress in human SH-SY5Y cells [3]. Prenylflavone derivatives from *B. papyrifera L.* were found to inhibit the

growth of breast cancer cells *in vitro* and *in vivo*, which showed that they had potent anti-tumor activity [4]. There are about 300,000 hectares of *B. papyrifera* in China, which are a potential resource of animal feeding with economical and practical merits. Many of the plants contain phytochemicals, which have potent antioxidant activities [1,5-8]. Antioxidant activities have been described for related polyphenolic constituents extracted from the stem, bark and wood of *B. papyrifera L.* [9]. It is reported that intake of high total antioxidant capacity (TAC) plants might be beneficial for the TAC status of cattle, and mean TAC levels in shrubs and trees were higher than those in grass, concentrate and timothy hay [10,11]. It has been suggested that flavonoids, a major class of plant secondary metabolites with many functions, such as pigmentation, antimicrobial activity and antioxidant activity [12], widely present in the plant kingdom, play an important role in disease prevention [13]. Oxidative stress has no clinical symptoms but is particularly dangerous in animals. Some defense mechanisms are available to prevent oxidative damage, including some enzymes, glutathione peroxidase (GSH-Px), superoxide dismutase (SOD), TAC, and catalase (CAT). Dietary antioxidants are vital components in preventing damage by free radicals in the body, like reactive oxygen species [14,15]. The objective of this study was to investigate the effects of *B. papyrifera* silage on the dry matter intake, blood plasma metabolites, immune function, antioxidant capacity and milk fatty acid (FA) profile in dairy cattle and to provide practical evidence supporting the application of *B. papyrifera* silage in the diet of dairy cows.

MATERIALS AND METHODS

The experimental protocol was approved by the Chinese Academy of Agricultural Sciences Animal Ethics Committee, which was performed in accordance with animal welfare practices and procedures followed the Guidelines for Experimental Animal of the Ministry of Science and Technology (2006, Beijing, China).

Animal and management

The whole foliage of *B. papyrifera* was mowed with 20 cm of stubble left from saplings of about 1.8 m height. The whole crop was ensiled without additive after chopping to a length of 2.5±1.5 cm, The *B. papyrifera* silage was fermented for 45 days before being used for the feeding experiment. Seventy-two Holstein cows with initial day in milk (DIM) = 128±65 d, body weight = 572±29 kg, milk yield = 32.16±0.95 kg and milk somatic cell counts (SCC) = $158\times10^3\pm35\times10^3$ cells/mL were divided into four groups randomly according to day in milk, parity, milk yield, and milk SCC, and allocated in 6 pens with 3 individuals in each group and given *ad libitum* access to feed and water. The cattle in the control group were fed on the original total mixed ratio (TMR) in the dairy farm, and the other 3 treatment groups were fed on the new TMR with 5% (Group low [Group L]), 10% (Group middle [Group M]), or 15% (Group high [Group H]) of *B. papyrifera* silage preparation, separately. The four different TMRs were adjusted as isonitrogenous and isoenergetic diets, with the composition shown in Table 2. The cattle were fed approximately 110% of expected consumption at 8:00 and 17:00 separately, and the residual was collected before the next feeding and then weighed and recorded. The delivery amount was adjusted according to the previous day's consumption. Cows were milked at 6:00, 13:00, and 19:30 in the milking parlor. The feeding adaption period was 7 days, and the experiment period was 42 days. The body condition score was measured by 2 evaluators and scores were averaged.

Measurements and analysis

The composited dietary samples, including feeding ingredients, TMRs and residual, were collected on fresh matter basis every 2 weeks during the experimental period. The samples were dried in an oven at 65°C for 48 h, then kept at room temperature for 24 h, and ground to pass through 40-mesh sieve. Dry matter (DM), crude fat (ether extract), ash, neutral detergent fiber (NDF), acid detergent fiber (ADF) were determined according to the methods reported by van Soest et al [16] and Goering and Van Soest [17], respectively.

The pH value of *B. papyrifera* silage was 5.04 and the nutrient composition of *B. papyrifera* silage was 29.06% of DM, 15.01% of crude protein (CP), 58.78% of NDF, 32.15% of ADF, 6.17% of ash at DM basis (Table 1), and 4 different TMRs are shown in Table 2. Milk yield was recorded every day, and milk samples were collected weekly, 40 mL of the milk was added with Potassium Dichromate for preservation and kept in –4°C for milk component analysis. Protein, fat and lactose concentrations in milk were determined with a Foss MilkoScanFT 6000 Analyzer (Foss Electric A/S, Hillerod, Denmark), and SCC were determined with a Foss Matic FM5000 instrument (Foss Electric, Denmark).

Another 50 mL of milk was frozen at –20°C without preservative for FA profile analysis. Total lipids in milk were extracted according to the method reported by Bligh and Dyer [18], then FAs were methylated into fatty acid methyl esters (FAME) according to the method described by Carrapiso et al [19]. The FA profile was determined by gas chromatography (GC, Shimadzu GC-2010, Kyoto, Japan) equipped with flame ion-

Table 1. The pH value and nutrient composition of *Broussonetia papyrifera* silage

pH value	DM	CP (% DM)	NDF (% DM)	ADF (% DM)	Ash (% DM)
5.04	29.06	15.01	58.78	32.15	6.17

DM, dry matter; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber.

Table 2. Nutrient composition of the diets

Item	Control	Group L	Group M	Group H
Ingredients (% of DM basis)				
B. papyrifera silage	0	5	10	15
Whole corn silage	23.99	16.65	10.34	4.03
Alfalfa hay	15.15	11.56	8.01	4.46
Oat hay	4.54	10.47	15.33	20.19
Beet pulp pellet	3.80	3.80	3.80	3.80
Whole cottonseed	7.60	7.60	7.60	7.60
Corn	22.05	22.05	22.05	22.05
Soybean meal	7.75	7.75	7.75	7.75
Wheat bran	3.80	3.80	3.80	3.80
Cottonseed meal	1.8	1.8	1.8	1.8
Rapeseed mean	3.0	3.0	3.0	3.0
DDGS	4.5	4.5	4.5	4.5
$NaHCO_3$	0.6	0.6	0.6	0.6
Limestone	0.6	0.6	0.6	0.6
$CaHPO_4{\cdot}2H_2O$	0.30	0.30	0.30	0.30
NaCl	0.22	0.22	0.22	0.22
Premix[1)]	0.3	0.3	0.3	0.3
Total	100	100	100	100
Chemical composition				
DM (%)	52.81	51.05	50.92	50.66
CP (%)	15.71	15.84	15.60	15.94
NE_L (MJ/kg of DM)[2)]	7.46	7.52	7.55	7.48
EE (%)	2.18	2.74	2.11	2.29
NDF (%)	29.08	28.95	29.69	28.87
ADF (%)	17.73	19.44	18.55	17.98
Ash (%)	7.29	7.85	7.45	7.46

DDGS, distillers dried grains with solubles; DM, dry matter; CP, crude protein; NE_L, net energy-lactation; EE, ether extract; NDF, neutral detergent fiber; ADF, acid detergent fiber.
[1)] The premix contains the following: Vit A 600,000 IU; Vit D 100,000 IU; Vit E 4,000 IU; Fe 3,000 mg; Cu 2,000 mg; Mn 2,500 mg; Zn 8,000 mg; Se 60 mg; I 100 mg; Co 20 mg per kilogram.
[2)] NE_L was a calculated value, while the others were measured values.

ization detector and split injection, 100 m length, 0.25 mm i.d., 0.2 μm thickness capillary GC column (SP-2560; Supelco Inc., Bellefonte, PA, USA).The initial oven temperature was 100°C, which was held for 13 min, then increased to 180°C at a rate of 10°C and held for 6 min, then increased to 200°C at 1°C/min and held for 20 min, and then increased to 230 at 4°C/min and held for 15.5 min. High purity nitrogen was used as carrier gas at a flow rate of 1 mL/min. The injector was set at 270°C, and the detector was set at 280°C. FAMEs were identified by compared with the retention times of the standards. The standards were FAME MixC4-C24 Unsatures (Sigma 18919, Germany).

Six blood samples (10 mL) were collected into lithium heparin Vacutainer tubes (Becton, Dickinsonand Company, Franklin Lakes, NJ, USA) using jugular venipuncture before feeding on d 4 during each week. Samples were centrifuged at 3,500 rpm for 20 minutes; the serum was aspirated and kept at −20°C for analysis of metabolites, immune and antioxidation capacity. Metabolite, immune parameters and anti-oxidation biomarkers were determined by a biochemical auto-analyzer (Hitachi automatic biochemical analyzer 7600, Tokyo, Japan) using commercially available kits: glucose (GLU), total protein (TP), albumin (ALB), globulin (GLB), non-esterified fatty acid (NEFA), blood urea nitrogen (BUN), immunoglobulin A (IgA), IgG, IgM, CAT, SOD, GSH-Px, TAC, and malondialdehyde (MDA) according to the manufacturer's instructions. The activity of tumor necrosis factor α were measured by a micro plate reader (BioTek ELX800, BioTek, Winooski, VT, USA) with commercially available kits.

Statistical analysis

The data were compared using one-way analysis of variance with Duncan's multiple comparisons. Significance was set at $p<0.05$ using SAS version 9.2 (SAS Institute Inc., Gary, NC, USA), and the data were expressed as mean and accompanied by standard error of the mean.

RESULTS

Milk yield and composition

As shown in Table 3, cows fed on diets with *B. papyrifera* silage had lower dry matter intake (DMI) than those fed on

Table 3. Effects of *Broussonetia papyrifera* silage on DMI, feed efficiency, milk yield and milk composition

Item	Treatment				SEM	p-value
	Control	Group L	Group M	Group H		
DMI (kg/d)	22.27[a]	21.31[b]	19.93[c]	18.41[d]	0.167	<0.0001
body condition score	3.52	3.36	3.63	3.47	0.180	0.753
Milk yield (kg/d)	32.65	32.05	31.76	30.74	0.278	0.098
Feed efficiency	1.46	1.52	1.59	1.67	0.030	0.318
Milk fat (%)	3.64	3.72	3.87	3.91	0.042	0.093
Milk protein (%)	3.32	3.30	3.34	3.23	0.023	0.333
Milk lactose (%)	4.99	4.91	5.01	5.02	0.011	0.132
Milk SCC (10^3 cells/mL)	183.83[a]	151.87[a]	105.23[b]	81.74[c]	10.330	0.003

DMI, dry matter intake; SEM, standard error of the mean; SCC, somatic cell count.
[a-c] Means within row with different superscripts differ significantly ($p<0.05$).

diet without *B. papyrifera* silage (p<0.05), and cows had lower DMI as the *B. papyrifera* silage increased in the diet (p<0.05). Cows had similar feed efficiency in control group and treatment groups (p>0.05). There was no difference between groups in milk yield, milk fat, protein and lactose (p>0.05), while cows in Group M and Group H had significantly lower milk SCC than that in control group and Group L (p<0.05).

Blood metabolites, immune, and antioxidant functions

Parameters of serum are presented in Table 4. No significant differences were found in TP, ALB, GLB, NEFA, BUN, GLU, GSH-Px, MDA (p>0.05) between groups. Compared with the control, 10% and 15% of *B. papyriferasilage* significantly increased the concentration of IgA and IgG (p<0.05), and 15% of *B. papyrifera* silage significantly increased the concentration of IgM (p<0.05), and 15% of *B. papyrifera* silage significantly increased CAT, SOD, and TAC in the serum (p<0.05). But serum CAT, SOD, and TAC activity of cows was not affected by the diet with 10% or 15% *B. papyrifera* silage.

Milk fatty acid profiles

Compared with the control, 15% of *B. papyrifera* silage diets significantly decreased C4:0 and C17:0 content in milk fat (p< 0.05), increased C18:1n9c and polyunstaturated fatty acid (PUFA) concentration in the milk (p<0.05) as shown in Table 5.

DISCUSSION

Milk yield and composition

DMI of cattle in the experiment decreased significantly (p< 0.05) as the content of *B. papyrifera* silage increased in the diet. The palatability of *B. papyrifera* silage might not be as good as the hay silage or flavonoids exceeded a certain level which affected the DMI of the cattle in the Group L, Group M, and Group H. However, with a lower DMI of cattle fed with *B. papyrifera* silage, the feed efficiency and milk yield were not affected (p>0.05). The SCC in milk is an indicator reflecting mammary health and milk quality. The physiological level of SCC in bulk cow milk is between 50×10^3 and 100×10^3 cells per mL, whereas 200×10^3 cells per mL is taken as the threshold to define a mammary quarter between healthy and infected [20,21]. In the present study, Group M and Group H had lower milk SCC than that of the control Group and Group L, which might due to the anti-inflammatory effect of the active ingredients in *B. papyrifera* [22-24]. It is reported that high content of milk protein was found in high SCC milk [25] and fat content was significantly increased from 3.55% to 3.72% by an increment of SCC from 100 to 200 cells/mL [26], but a decrease of milk protein content when SCC in milk was high [5]. Lactose content in high-SCC milk was significantly lower [27] which could be a result of reduced synthesis because of the inflammation processes. In our study, no significant differ-

Table 4. Effects of *Broussonetia papyrifera* silage silage on serum indexes

Item	Treatment				SEM	p-value
	Control	Group L	Group M	Group H		
Serum metabolites						
TP (g/L)	57.98	59.32	56.97	54.87	2.153	0.915
ALB (g/L)	22.07	21.75	21.22	20.55	0.618	0.852
GLB (g/L)	35.92	37.57	35.75	34.32	1.663	0.933
NEFA (μmol/L)	445.64	386.80	480.29	369.18	31.585	0.601
BUN (mmol/L)	3.87	3.55	3.44	4.10	0.138	0.329
GLU (mmol/L)	3.38	3.72	2.83	2.56	0.268	0.438
Immune parameters						
IgA (g/L)	0.82^{b}	0.80^{b}	0.99^{a}	1.00^{a}	0.029	0.008
IgG (g/L)	8.29^{b}	9.23^{ab}	9.75^{a}	10.16^{a}	0.218	0.013
IgM (g/L)	0.75^{b}	0.77^{b}	0.81^{ab}	0.86^{a}	0.014	0.017
TNFα (ng/mL)	1.67	1.89	1.93	2.00	0.063	0.357
Anti-oxidation biomarkers						
CAT (U/mL)	3.96^{b}	4.26^{b}	4.80^{ab}	5.77^{a}	0.209	0.008
SOD (U/mL)	136.73^{b}	140.98^{b}	141.83^{ab}	152.47^{a}	2.099	0.041
TAC (U/ml)	9.25^{b}	10.58^{ab}	10.71^{a}	11.39^{a}	0.271	0.024
GSH-Px (U/mL)	844.72	886.18	856.10	848.78	10.107	0.486
8-OHdG (ng/mL)	0.49^{a}	0.44^{ab}	0.39^{bc}	0.33^{c}	0.017	0.001
MDA (μmol/L)	5.88	5.65	5.03	5.19	0.205	0.437

SEM, standard error of the mean; TP, total protein; ALB, albumin; GLB, globulin; NEFA, non-esterified fatty acid; BUN, urea nitrogen; GLU, glucose; IgG, immunoglobulin G; IgA, immunoglobulin A; IgM, immunoglobulin M; TNFα, tumor necrosis factor α; CAT, catalase; SOD, superoxide dismutase; TAC, total antioxidant capacity; GSH-Px, glutathione peroxidase; 8-OHdG, 8-hydroxy-2′-deoxyguanosine; MDA, malondialdehyde.
[a-c] Means within row with different superscripts differ significantly (p < 0.05).

Table 5. Effects of *Broussonetia papyrifera* silage on milk fatty acid profiles

Item (mg/g total lipid)	Treatment				SEM	p-value
	Control	Group L	Group M	Group H		
C4:0	1.39[a]	1.25[a]	1.08[b]	0.86[c]	0.046	<0.0001
C6:0	1.49	1.31	1.34	1.47	0.031	0.161
C8:0	1.16	1.13	1.09	1.07	0.024	0.755
C10:0	3.27	3.26	3.05	2.95	0.076	0.540
C12:0	4.29	4.22	3.83	3.63	0.104	0.057
C13:0	0.27	0.30	0.27	0.21	0.012	0.109
C14:0	14.58	14.90	14.66	13.95	0.190	0.364
C14:1	1.21	1.16	1.15	0.86	0.064	0.196
C15:0	1.49	1.50	1.54	1.34	0.045	0.434
C16:0	39.04	39.82	39.27	38.35	0.482	0.090
C16:1	1.76	1.56	1.61	1.53	0.072	0.753
C17:0	0.79[a]	0.70[a]	0.61[ab]	0.47[b]	0.041	0.026
C18:0	10.28	9.53	10.25	11.63	0.418	0.119
C18:1n9c	15.09[b]	15.29[b]	15.82[ab]	17.12[a]	0.295	0.042
C18:2n6c	2.81	2.90	3.22	3.39	0.096	0.138
C18:3n3	0.30	0.32	0.39	0.32	0.017	0.256
C20:3n6	0.17	0.18	0.19	0.18	0.014	0.489
C20:4n6	0.23	0.22	0.28	0.27	0.011	0.151
C21:0	0.38	0.45	0.35	0.40	0.016	0.224
SFA (% total FA)	78.43	78.37	77.34	76.33	0.382	0.057
UFA (% total FA)	21.57	21.63	22.66	23.67	0.353	0.203
PUFA (% total FA)	3.51[b]	3.62[ab]	4.08[a]	4.16[a]	0.103	0.025
MUFA (% total FA)	18.06	18.01	18.58	19.51	0.328	0.226

SEM, standard error of the mean; SFA, saturated fatty acid; FA, fatty acid; UFA, unsaturated fatty acid; PUFA, polyunstaturated fatty acid; MUFA, monounsaturated fatty acid.
[a-c] Means within row with different superscripts differ significantly ($p < 0.05$).

ences in milkfat, protein and lactose was found in difference levels of milk SCC which is consistent with the result reported by Mazal et al [28]. There might be no connection between the SCC and protein content in milk when the udders of cows are not infected.

Blood metabolites, immune and antioxidant functions

Blood metabolites represent an integrated index of nutrient supply in relation to the utilization of nutrients. In the present study, no significant difference was found in the serum TP, ALB, GLB, NEFA, BUN, and GLU between groups ($p>0.05$). The concentration of TP, ALB, and ALB were in the normal range, which indicates that the animals were in healthy status with no adverse effect on their performance [29]. The concentration of plasma GLU, NEFA, and BUN are directly related to timing of feed intake. NEFA is an indicator of energy balance and increased NEFA indicates lipid mobilization to meet energy demands, and intake time of protein, especially rumen undegraded protein. No significant differences were found in the concentrations of NEFA, BUN, and GLU in cows between groups, which probably demonstrates no potential metabolic changes from being fed on the amount of *B. papyrifera* silage. The immune system is a complex system regulating group of cells with integrated function essential to health. It is reported that plant flavonoids modulate immune and inflammatory cell functions [30].

In the present study, cows in Group M and Group H had significantly higher IgA and IgG than those in control group and cows in Group H had higher IgM than those in control Group, which might be due to the flavonoids in the *B. papyrifera* silage. Oxidative stress, an imbalance between free radical generation and antioxidant system, is a main contributing factor inducing various diseases in animals [31]. Antioxidant nutrients could be used to reduce the effects of substances that induce reactive oxygen metabolites and to control oxidative stress [32]. Some specific photochemicals isolated from the fruit of *B. papyrifera* have potent antioxidant effects [1]. Flavonoids are a major class of plant secondary metabolites with multiple functions, such as pigmentation, antimicrobial activity and antioxidant activity [12]. It is reported that they are naturally occurring in bark [33], fruit [15] and leaves [34] of *B. papyrifera*. SOD is the first line of antioxidant defense that catalysis the conversion of superoxide radical to hydrogen peroxide [35], while CAT is another antioxidant enzyme that can catabolize peroxide to water [36]. 8-Hydroxy-2'-deoxyguanosine (8-OHdG) is believed to be one of the most abundant DNA lesions resulting from oxidative stress and is a biomarker of the oxidative DNA damage and repair. The diet

with 15% of *B. papyrifera* silage significantly increased concentration of CAT, SOD, and TAC and decreased the serum concentration of 8-OHdG. Phenolic compounds and flavonoids appear to be the main components responsible for the antioxidant activity of *B. papyrifera* silage, and the antioxidant capacity was improved as the supplement level of *B. papyrifera* silage increased. The efficiency of antioxidative systems against reactive oxygen species is monitored by the estimation of TAC. It is reported that mean TAC levels in shrubs and trees were higher than those in grass, concentrate and timothy hay, and intake of high TAC plants might be beneficial for the TAC status of cattle [10,11]. The MDA is a final product of lipid peroxidation. Cows in Group H had significantly higher TAC than that in control Group, but there was no significant difference between the groups, which is in accordance with results that an increase in TAC is not parallel with an increase in MDA in cows [37]. Supplementation of *B. papyrifera* silage did not affect the serum concentrations of MDA in dairy cows, which might due to the length of the experiment or the dosage of *B. papyrifera* silage not being in an appropriate range. Some non-enzymatic antioxidants, like GSH-Px, α-tocopherol, *β*-carotene and uric acid represent the primary antioxidant capacity of serum [35] and could be used as a tool to evaluate the general nutritional status of animals [38]. In the present study, nutrition didn't appear to have significant effect on the activity of GSH-Px in blood which is in accordance with the studies by Sgorlon et al [39], and it might be sensitive at tissue or cellular level.

Milk fatty acid profiles

Because of the biohydrogenation occurring in the rumen, milk and other dairy products are a major contributor to dietary saturated fatty acid, which has led to a negative consumer perception and a public health concern related to excessive intake of saturated fats [40]. There have been numerous researches carried out during the last three decades to improve the FA profiles of the products of ruminants [41-44], and there was not enough evidence of direct relationship between PUFA and health, however, further studies indicated PUFA content of the foods did affect some indicators of health status [45-47]. Compared with the control group, milk PUFA of cows are significantly higher ($p<0.05$), and saturated fats (C4:0, C17:0) are significantly lower ($p<0.05$) in group M and group H, which might be caused by the reduced biohydrogenation of unsaturated fatty acids in the rumen. It is reported that the active ingredients of *B. papyrifera* have antibacterial or antimicrobial effect [33], and this might have affected the rumen microflora, which reduced the hydrogenation of FA in the rumen. Dietary PUFA act as precursors for inflammation-mediating eicosanoids and resolvins, and represent direct ligands of transcription factors associated to immune response [48-50]. With more PUFA, the milk would be more beneficial for human's health.

CONCLUSION

B. papyrifera silage could be used as a new feed resource for lactating dairy cows, affecting their antioxidant capacity and the milk components positively. Supplementation with about 10% to 15% of *B. papyrifera* silage might reduce milk SCC, while improving the quality of milk with more PUFA, and improve the immune functions and the antioxidant functions. It could serve as a new feeding resource for silage preparation. Phenolic and flavonoids might be the main components responsible for the antioxidant activity in the *B. papyrifera* silage which needs further study. Moreover, further investigation is required to investigate the mechanism in which the *B. papyrifera* silage regulates the immune functions and antioxidant functions of dairy cows.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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