Original Research Article

Phytonutrient pellet supplementation enhanced rumen fermentation efficiency and milk production of lactating Holstein-Friesian crossbred cows

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A B S T R A C T

The objective of this experiment was to investigate the effects of inclusion of dragon fruit peel pellet (DFPP) and dietary non-protein nitrogen (NPN) on nutrients digestibility, rumen fermentation efficiency, plasma antioxidant activity, microbial protein synthesis, milk yield and composition in lactating Holstein-Friesian crossbred cows. Four animals were randomly allotted to 4 dietary treatments according to a $2^2$ factorial arrangement in a $4 	imes 4$ Latin square design. The treatments were as follows: 300 g DM of DFPP + 100 g of urea (T1), 300 g DM of DFPP + 200 g of urea (T2), 400 g DM of DFPP + 100 g of urea (T3), and 400 g DM of DFPP + 200 g of urea (T4), respectively. The results showed that intake of rice straw was increased ($P < 0.01$) by the DFPP addition. Including DFPP and urea did not affect ($P > 0.05$) the NDF and ADF digestibilities, but increased the apparent digestibilities of dry matter, organic matter, and crude protein ($P < 0.01$). Rumen fermentation process, especially the propionate concentration, was significantly increased by the DFPP levels. The plasma antioxidant activity was increased ($P < 0.05$) with the addition of DFPP. The DFPP improved ($P < 0.01$) microbial protein synthesis. The supplementation of DFPP and urea increased ($P < 0.05$) milk fat, whereas milk yield and 3.5% fat corrected milk were only increased ($P < 0.05$) by the DFPP supplementation. Based on these results, addition of DFPP at 400 g/animal per day improved rumen fermentation, plasma antioxidant activity, milk yield and milk fat percentage.

1. Introduction

Industrial fruit processing has been increasingly important to avail fruit-products for human consumption. The use of fruit-wastes has been receiving more attention as they contain phytonutrients (PTN) sources (Ibrahim et al., 2017). Phytonutrients are a major group of secondary metabolites commonly found in fruit peels and wastes especially the phenolic compounds (PC), condensed tannins (CT), and saponins (SP), which play a vital role in animal health and nutrition (Abbas et al., 2017). Furthermore, these compounds exert impacts on antioxidant activity which can prevent the availability of free radicals (Borges et al., 2010). Free radicals and reactive oxygen species are continuously formed in the body of animal (Aruoma, 1998). Generally, animals are naturally protected against reactive oxygen species or free radicals via various natural antioxidant enzymes. However, animals from tropical areas are inclined to oxidative stress due to prolonged exposure to high temperatures (Salles et al., 2010). In ruminant feeding, there has been a growth of interest in the study of oxidative stress determined as an imbalance between oxidants and antioxidants to understand the function of oxidant and antioxidant molecules in physiological conditions (Lykkesfeldt and Svendsen, 2007).

Dragon fruit (Hylocereus undatus) peel is one of the alternative sources of plant containing antioxidants, especially PC that is non-

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toxic and biologically safe. A current study has shown that antioxidant addition in the ruminant diet could alleviate oxidative stress status (Rizzo et al., 2013) and increase plasma antioxidant capacity (Tian et al., 2019). Moreover, these plants contain phytonutrients such as CT and SP, with the specific effect of improving milk profile, nitrogen efficiency in dairy cows (Frutos et al., 2004), enhancing rumen fermentation (Kang et al., 2014; Cherdthong et al., 2019), as well as a protective effect on ruminal protein in order to promote duodenal utilization and modify the volatile fatty acid pattern in the rumen (Salem et al., 2014). However, dietary non-protein nitrogen (NPN) supplementation has been shown to enhance the crude protein level of low quality agro-industrial by-products (Foiklang et al., 2016). Nevertheless, not much research has been carried out on the use of dragon fruit peel and NPN in vivo trials especially in lactating dairy cows.

Hence, this present experiment investigates the effect of dragon fruit peel pellet (DFPP) as a source of PTN in combination with NPN on nutrient digestibility, rumen fermentation efficiency, plasma antioxidant capacity, oxidative stress, microbial protein synthesis, milk yield and composition in lactating Holstein-Friesian crossbred cows.

2. Materials and methods

2.1. Animal care and management

All Holstein-Friesian crossbred cows were approved by the Institute of Animals for Scientific Purpose Development (IAD) of Thailand (protocol no. U1-06878-2560). Dairy cows were injected with vitamin AD3E and drenched with ivermectin (1 mL per 50 kg live weight) before imposing the respective treatments. Animals with vitamin AD3E and drenched with ivermectin (1 mL per 50 kg live weight) before imposing the respective treatments. Animals were housed in a ventilated barn and in individual pens (3 m × 4 m). Mineral blocks and clean drinking water were available freely.

2.2. Dragon fruit peel pellet preparation

The fresh peels were obtained from a fruit canning factory in Thailand. The fruit peels were dried in an oven at 60 °C for approximately 3 d and were ground (Cyclotech Mill, Tecator, Höganäs, Sweden) then the dragon fruit powder was mixed with ingredients (Table 1) to pellet form by using a Ryuozo-kun pelleting machine (Kakiuchi Co., Ltd., Nankoku, Kochi, Japan).

2.3. Experimental procedure

Four Holstein-Friesian crossbred cows were used in a 2 × 2 factorial in a 4 × 4 Latin square design. Pre-experimental live weights were 525 ± 25 kg, 123 ± 21 d-in-milk (DIM), and an average milk yield of 13.8 ± 1.8 kg/animal per day. The study lasted for 84 d, with four 21-d experimental periods; the first 14-d for adaptation to dietary treatments and last 7 d for sample collection. Animals were fed concentrate to milk yield ratio at 1:1.5 and rice straw was fed at free choice. Cows were hand milked at 06:00 and 16:00.

2.4. Dietary treatments

The dietary treatments were two factors of DFPP, fed at 300 and 400 g DM/animal per day, and urea at 100 and 200 g/animal per day. Thus, the factorial arrangement of treatments were as follows: 300 g DM of DFPP + 100 g of urea (T1), 300 g DM of DFPP + 200 g of urea (T2), 400 g DM of DFPP + 100 g of urea (T3), and 400 g DM of DFPP + 200 g of urea (T4), respectively. The DFPP and urea supplementation were top-dressed on the concentrate, manually mixed and fed to animals.

2.5. Chemical composition of feeds

The DFPP and concentrate mixture were prepared following the respective ingredients shown in Table 1. The concentrate mixture (14% CP and 78% TDN) containing 55.0% cassava chip, 19.0% soybean meal, 10.5% dried brewer’s grains, 10.0% rice bran meal, 4.0% molasses, 1.0% mineral premix, 0.5% sulfur was then mixed well. The dry matter (DM; No. 967.03), ash (No. 942.05), crude protein (CP; No. 984.13) in the samples were chemically analyzed by using the method of AOAC (2012; No. 973.18), and for CT following the modified method

| Item | Concentrate | Rice straw | Dragon fruit peel powder | Dragon fruit peel pellet |
|------|-------------|------------|--------------------------|------------------------|
| Ingredients, % as fed | | | | |
| Cassava chip | 55.0 | – | – | – |
| Soybean meal | 19.0 | – | – | – |
| Dried brewers’ grains | 10.5 | – | – | – |
| Rice bran meal | 10.0 | – | – | – |
| Molasses | 4.0 | – | 1.0 | 1.0 |
| Sulfur | 0.5 | – | 1.0 | 1.0 |
| Mineral premix | 0.5 | – | 1.0 | 1.0 |
| Urea | – | – | 1.0 | 1.0 |
| Salt | – | – | 1.0 | 1.0 |
| Dragon fruit peel powder | – | – | 90.0 | 90.0 |
| Cassava starch | – | – | 5.0 | 5.0 |
| Chemical composition, % dry matter | | | | |
| Dry matter, % as fed | 87.7 | 90.6 | 90.4 | 86.5 |
| Ash | 4.9 | 12.3 | 1.5 | 1.4 |
| Crude protein | 13.9 | 3.0 | 5.4 | 9.7 |
| Neutral-detergent fiber | 17.5 | 71.3 | 33.0 | 17.8 |
| Acid-detergent fiber | 10.1 | 55.5 | 29.6 | 14.5 |
| Condensed tannins | – | – | 6.9 | 6.2 |
| Saponins | – | – | 8.9 | 8.1 |
| Total phenolic content, mg/100 g | – | – | 35.4 | 34.2 |

1 One kilogram of mineral premix contains the following content: vitamin A 10,000,000 IU; vitamin D 1,800,000 IU; vitamin E 70,000 IU; Fe 50 g; Mn 40 g; Zn 40 g; Cu 10 g; I 0.5 g; Se 0.1 g; Co 0.1 g.
of vanillin-HCl as described by Wanapat and Pongchompu (2001). Total phenolic content (TPC) was determined by the modification of the Folin-Ciocalteu spectrophotometric method (Lim et al., 2006).

2.6. Sample collection and chemical analyses

Feed refusals were recorded daily before the morning feeding and dry matter consumption of each animal was measured. Feces, urine, and milk samples were collected during the last 7 d of each experimental period. Rumen fluid (0 and 4 h after feeding) was sampled from cows, on the last day of each period and taken through a tube, connected with a vacuum pump inserted via the mouth to the middle part of the rumen and into a plastic beaker. On the last day, blood samples 10 mL (0 and 4 h-after-feeding) were collected (from the jugular vein) and 12-mg ethylenediamine tetraacetic acid (EDTA) was then added.

Approximately 5% of total fresh fecal samples were collected daily and separated into two portions; the first portion of each day was analyzed for DM (No. 967.03) content (AOAC, 2012; No. 973.18) and the last portion was pooled at the end of each period and for each animal. The pooled fecal samples (500 g) were stored at 20 °C until later analysis for other compositions. The urine samples were corrected at 10% of total output from each animal. Individual urine (45 mL) was homogenized with 1 mol/L sulfuric acid (5 mL) to prevent volatilization of ammonia and frozen to −20 °C until later analysis. The urinary allantoin was analyzed by using the method of Chen et al. (1993). The concentration of microbial purine derivatives (PD) absorption (X; mmol/d) corresponding to the PD excretion (Y; mmol/d) was calculated based on their relationship following to Chen and Gomes (1992): Y = 0.85X + (0.385BW0.75). The microbial N supply (MNS) was estimated by using the method of Chen and Gomes (1992): MNS (g/d) = (X × 70)/(0.116 × 0.83 × 1,000) = 0.727 × X, where X is PD absorption (mmol/d). The ratio of protein-N to total N in mixed rumen microbes is 11.6:100. The microbial protein digestibility is 0.83 and the N concentration of protein is 70 mg N/mmol. The efficiency of microbial nitrogen synthesis (EMNS) was based on the equation defined by Chen and Gomes (1992): EMNS = microbial nitrogen (g/d)/DOMR, where DOMR is digestible organic matter fermented in the rumen (the ruminal digestion was 650 g/kg OM of digestion in total tract). DOMR = DOMI × 0.65, where DOMI is digestible organic matter intake, which followed the assumptions calculated by ARC (1984).

One of the milk samples was combined following the proportion of milk production at milking time (using a ratio of the morning to afternoon at 60:40) and preserved with potassium dichromate (K2Cr2O7) then stored at 4 °C until later analysis for compositions. Milk samples were analyzed for content of protein, fat, solids-not-fat (SNF), total solids (TS), lactose, galactose, glucose, casein, and milk urea nitrogen (MUN) using MilkoScan FT1 (FOSS analytical A/S, Hillerod, Denmark).

After discarding the first 200 mL of rumen fluid to minimize saliva contamination, approximately 50 mL of a sample of rumen fluid was collected. Rumen fluid was immediately detected for pH using a pH meter (HANNA Instruments HI 8424 microcomputer, Singapore). Samples (45 mL) were mixed with 1 mol/L sulphuric acid (5 mL) to stop the microbe activity and then centrifuged (3,000 e 20) and stored at −20 °C until analysis concentration of volatile fatty acids (VFA) (Samuel et al., 1997); using high-performance liquid chromatography (HPLC) (Model Water 600; UV detector, Millipore Crop) and ammonia nitrogen (NH3-N) (AOAC, 2012; No. 973.18); using kjeldahl process (Kjeltech Auto 1030 analyzer, Tecator, Hoganas, Sweden). Rumen samples (1 mL) and 10% formalin (9-mL) were mixed well for the total direct counts of bacteria and protozoa (Galyean, 1989), which were calculated using a haemocytometer (Boeco, Hamburg, Germany). Details of the experimental protocols were stated in Wanapat et al. (2014).

The blood samples were centrifuged at 500 × g for 10 min at 4 °C to separate plasma and stored at −20 °C for blood urea nitrogen (BUN) analysis (Croker, 1967). Furthermore, the lipid oxidation of plasma was determined with thiobarbituric acid reactive substances (TBARS) (Toaldo et al., 2015). Briefly, supernatants were measured with absorbance at 532 nm via UV absorption spectrophotometry (T80 + UV/Vis, PG Instruments Ltd., Leicestershire, United Kingdom). The results were calculated as TBARS concentration expressed in μmol/L of malondialdehyde (MDA) using the molar extinction coefficient of the pink TBA chromagen as 1.56 × 10⁻⁵. The antioxidant capacity was determined spectrophotometrically according to the method of Martinez et al. (2006), using a stable free radical 1-diphenyl-2-picrylhydrazyl (DPPH). The absorbance was determined at 517 nm using a UV–Vis spectrophotometer (SpectraMax M3 Multi-Mode Microplate Reader, Molecular Devices, San Jose, CA, USA).

2.7. Statistical analyses

The data variables were analyzed according to a 4 × 4 Latin square design with a 2 × 2 factorial arrangement (two levels of DFPP and NPN). All parameters were analyzed by ANOVA using the general linear model (GLM) procedure (SAS, 2013). Treatment effects were tested using Duncan’s new multiple range test. Significant effects were identified at P < 0.01 and P < 0.05.

3. Results

3.1. Nutrient composition of diets

Ingredients and chemical composition of the diets are presented in Table 1. The concentrate used a cassava chip as an energy source and contained CP, NDF, and ADF at 13.9%, 17.5%, and 10.1% DM. The rice straw consisted of 3.0% CP, 71.3% NDF, and 55.5% ADF, and DFPP contained 9.7% CP, 17.8% NDF, and 14.5% ADF, respectively. Importantly, DFPP contained phytonutrient compounds especially total PC at 34.2 mg/100 g, 8.1% SP, and 6.2% CT, respectively.

3.2. Dry matter consumption and nutrient digestibility

As shown in Table 2, the concentrate consumption was greater (P < 0.03) in animals fed with the levels of DFPP at 400 g/animal per day than those at 300 g/animal per day, and there was no difference in percent body weight by the DFPP and urea supplementation. The rice straw and total DM consumption were significantly enhanced (P < 0.001) with the DFPP supplementation. Digestibilities of DM, OM, and CP were significantly different among treatments (P < 0.001). The supplementation of DFPP and urea significantly increased (P < 0.001) CP digestibility, especially the addition of DFPP at 400 g/animal per day combined with urea at 200 g/animal per day. Moreover, no differences were found in fiber digestibilities (NDF and ADF) of the cows.

3.3. Rumen fermentation and blood metabolite

Rumen fermentation and blood metabolite parameters are shown in Table 3. The rumen pH was not influenced among treatments. The NH3-N concentration was higher (P < 0.001) in DFPP and urea supplemented with 400 and 200 g/animal per day. Rumen microbes, and total bacteria group were significantly increased (P < 0.001) with DFPP and urea supplementation, whilst the protozoa population was reduced (P < 0.001) when DFPP levels were
incremental. Furthermore, the addition of DFPP and urea were associated with differences ($P < 0.001$) in BUN concentration.

### 3.4. Rumen volatile fatty acids

The concentrations of acetate, propionate, acetate:propionate ratio, and total VFA in the rumen fluid of cows supplemented with DFPP were significantly enhanced ($P < 0.001$), whilst butyrate production was not impacted by the DFPP and urea supplementation. The proportion of propionate was increased ($P < 0.001$) with the level of DFPP at 400 g/animal per day (Table 3).

### 3.5. Plasma antioxidant activity and oxidative stress

As shown in Table 4, the supplementation of DFPP resulted in enhancements in DPPH scavenging activity measured at 0 and 4 h after feeding ($P < 0.001$), whilst butyrate production was not impacted by the DFPP and urea supplementation. The proportion of propionate was increased ($P < 0.001$) with the level of DFPP at 400 g/animal per day when compared with the group fed DFPP at 300 g/animal per day. MDA was not influenced between cows supplemented with the different treatments.

### 3.6. Urinary purine derivatives and microbial protein synthesis

Allantoin (absorption and excretion) concentrations showed higher significance ($P < 0.001$) with the DFPP and urea supplementation. Microbial nitrogen supply (MNS) and efficiency of microbial nitrogen supply (EMNS) were significantly increased when cows were supplemented with DFPP ($P < 0.001$) and urea ($P = 0.04$; Table 5).

### 3.7. Milk yield and composition

The data for milk yield and composition in cows supplemented with the DFPP and urea are taken in Table 6. Both milk production and 3.5% fat collected milk (kg/d) were greater ($P = 0.03$ and $P = 0.01$) when DFPP at 400 g/animal per day was added to the diet. Enhancement in milk fat was significantly higher ($P = 0.02$) when cows were fed DFPP and urea in the diet. In contrast, milk protein,
solid not fat, total solids, lactose, galactose, glucose, and casein were not significantly affected by the treatments. Moreover, no difference in MUN was observed in the group fed DFPP and urea.

4. Discussion

4.1. Dry matter consumption and nutrients digestibility

One important limitation in feeding CT and SP in ruminant feeds is the low palatability, which may limit the DM consumption of feeds below the levels expected (Wang et al., 2018). Additionally, when exceeding a CT concentration (approximately >6% DM in the diet), a reduction in feed consumption is commonly observed (Peng et al., 2016). Under this experiment, it was shown that moderate amounts of CT and SP in the DFPP might enhance digestion without affecting DM intake. These results agree with Avila et al. (2020) who stated that Acacia mearnsii fed at 20 g/kg of DM in the diet had no reduction in the dry matter intake.

In the present experiment, the addition of DFPP and urea to the diet clearly improved DM, OM, and CP digestibilities. This could be attributed to the increase of microbial population which would degrade more feeds. Moreover, this finding could be due to DFPP which contained CT and SP. The presence of phytonutrients improved the digestibilities of DM and OM, which could be due to the concentration used causing a fluctuating shift in the rumen microorganisms, thus did not impact feed digestion. The CP
digestibility was significantly increased by the DFPP addition; this change could be attributed to the binding of CT with protein which was able to increase rumen by-pass protein. This finding agrees with Wang et al. (2018) who reported that adding CT and SP often entails adverse effects on nutrient digestibility, especially protein digestion where a decreased rumen protein digestion is desirable, provided the tannin-CP bonds are later separated and the protein can be digested in the lower-gut. Ampapon et al. (2019) evaluated the use of phytonutrients based on using both mangosteen peel powder and banana flower powder and found the enhancement of fiber digestibility with increasing the rumen bacteria population.

### 4.2. Rumen fermentation and blood metabolite

The rumen pH of dairy cows was unaffected by the DFPP and urea levels, ranging from 6.6 to 6.7. This is a notable range that was suitable for fibrolytic bacteria population to breakdown of fiber in the rumen and with no impact on rumen acidosis, as demonstrated by Wanapat et al. (2014) and Abdela (2016). Accordingly, Dschaa et al. (2011) revealed that rumen pH was not influenced by level of CT in lactating dairy cows. Rumen pH is one of the most important functions of fermentation variables instantly affecting microbial activity and growth (Li et al., 2009).

The rumen NH₃-N concentration was improved by the DFPP and urea supplementation and was higher in DFPP supplemented with 200 g/animal per day of urea. This could be a positive effect of the DFPP which contained 5.4% CP and NPN, resulting in enhancement of the NH₃-N content. Supapong et al. (2017) explained that the concentrate, protein, and roughage sources in the diet that can affect the animals is based on the knowledge of which NH₃-N is the major product of rumen protein digestion. Most of the nitrogen used by rumen microorganisms was provided from the available NH₃-N pool in the rumen. Accordingly, adding DPFF at 400 g/animal per day to Holstein crossbred bulls resulted in increased rumen NH₃-N concentration (Matra et al., 2020).

The bacterial population was increased by the DFPP and urea levels, while the protozoal population was decreased. It is well known that PTN presents a broad range of anti-microbial effects against microbial populations, especially protozoa and PTN, attributed to the hydrophobicity of their active compounds (Calsamiglia et al., 2007; Patra, 2012). Perna Junior et al. (2017) used A. mearnsii containing CT at 6 g/kg in dry cows, which resulted in a decrease in the protozoal population. Moreover, SP-containing plants or extracts have inhibitory effects on protozoa as shown by Patra and Saxena (2009). In addition, Fagundes et al. (2020) reported that tannin-rich forage increased the rumen bacteria population.

The concentration of BUN was significantly improved in dairy cows given DFPP and urea. This may be because BUN correlated with the NH₃-N concentration availability in the rumen. BUN concentration could be employed to detect nitrogen metabolism in ruminants, as higher BUN can reflect higher protein degradation in the rumen (Broderick and Clayton, 1997).

### 4.3. Rumen volatile fatty acid

Under this investigation, the molar proportion of propionate and total VFA were significantly improved by DFPP supplementation, whereas the acetate proportion was decreased. This may be related to the effect of phytonutrients on bacterial population in the rumen. The increase in bacterial population in this trial could improve the rumen fermentation, thus yielding higher total VFA and propionate production. Accordingly, Ampapon and Wanapat (2020) stated that addition of rambutan peel powder at 4% dry matter intake subsequently increased rumen propionate production in beef cattle. Furthermore, Khiao-Ard and Zebeli (2013) showed that PTN was effectively improving propionate and reducing acetate concentration in dairy cows.

### 4.4. Plasma antioxidant activity and oxidative stress

The levels of DPPH scavenging activity were greater in animals fed with DFPP at 400 g/animal per day. This might be a positive effect of the DFPP containing PC. These compounds are effective as dietary antioxidants in animal feeding which has been receiving more attention. PC is a natural antioxidant that is ingested by animals and is believed to contribute towards an enhanced antioxidant activity (Vasta and Luciano, 2011). In agreement with these observations, López-Andrés et al. (2014) stated that PC present in ryegrass could contribute to enhancing the antioxidant activity of the plasma of grazing lambs. This important mediator of cellular macromolecule damage must be continuously eliminated and controlled by antioxidant mechanisms to prevent various diseases especially mastitis (Konvica et al., 2015). Antioxidants play an essential role against the deteriorating action of free radicals in the organisms. Insufficiency of antioxidants in living organisms leads to oxidative stress (Pourreza, 2013).

MDA is the end-product of lipid peroxidation, thus is used as an index of oxidative stress (Esterbauer et al., 1991). In this recent study, MDA was not influenced by DFPP supplementation. This could be due to PC impacting on antioxidant status, thus MDA could be maintained at a normal range (1.0 to 1.5). Similarly, Safari et al. (2018) showed that the plasma concentration of MDA was not influenced by pomegranate by-products in transition dairy cows.

### 4.5. Urinary purine derivatives and microbial protein synthesis

In the present study, estimated urinary purine derivatives were significantly increased with DFPP in combination with urea. Accordingly, lower urine allantoin excretion has been reported in cows fed a diet with lower CP percentage than those of present study Cutrignelli et al. (2007). This could be due to a higher level of DFPP supplementation, hence higher N intake, together with its effect on forming of protein complex. The high dietary CP intake increased the flow of protein from the rumen to the lower gut (Giang et al., 2016). The rumen microbial protein synthesis additionally plays an essential role in ruminants since it provides a high level of protein resources for host animals. Microbial protein synthesis efficiency depends on DM intake, rumen carbohydrate and the protein rate of fermentation, and rumen dilution rate (Stern et al., 1997). Furthermore, MNS and EMNS were significantly increased with DFPP and urea addition. Similarly, Viennasay and Wanapat (2020) found in lactating dairy cows that using Flemingia could improve MNS and EMNS. Production of microbial protein synthesis (MPS) using NH₃-N released from rumen protein degradation would support the outflow of protein into the small intestine (Russell and Rychlik, 2001).

### 4.6. Milk yield and composition

Milk yield and milk fat were increased when supplemented with DFPP. This could be due to the CT and SP present in the DFPP. Accordingly, Benchaar et al. (2008) reiterated that CT and SP when contained at moderate concentration could be beneficial to rumen fermentation end-products and production. Several studies have shown positive effects from tropical fruit peel by-products on rumen fermentation, as well as milk yield; using mangosteen peel pellet (Norrapoke et al., 2012) and rambutan peel powder (Ampapon and Wanapat, 2020). Importantly, high levels of propionate concentration would be converted into glucose via
gluconogenesis which could produce more NADPH via the pathway and be utilized later for the synthesis of fatty acids (Isobe et al., 2011).

5. Conclusions

DFPP was an alternative by-product resource containing phytonutrients namely PC, CT, and SP. Based on the findings, supplementation of DFPP at 400 g/animal per day with urea at 100 g/animal per day enhanced rumen fermentation characteristics, microbial protein synthesis, milk yield and milk fat, as well as increased plasma antioxidant activity. Therefore, DFPP could be a promising dietary rumen enhancer to replace chemicals and antibiotics for ruminant feeding.

Author contributions

Maharach Matra: Conceptualization, Methodology, Data collection, Formal analysis, Writing – Original draft preparation. Metha Wanapat: Conceptualization, Methodology, Writing – Original draft preparation, Reviewing and Editing.

Declaration of competing interest

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, and there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

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