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Effect of Cocoa Pulp Level Mixed With Feed Concentrate on Performance and Blood Metabolite Profiles of Dry-Lot Fattening Bali Steers

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Abstract: Aim of this study is to evaluate the utilization of cocoa pulp as an alternative feedstuff source for the performance and blood metabolite profiles of dry-lot fattening Bali steers. Nine fattening Bali steers were randomly assigned to experimental diets, composed of three levels of cocoa pulp of 0, 5 and 10% with reducing levels of milled corn composition at 20, 14 and 8%, designated as ED0 (control), ED1 and ED2, respectively, under a completely randomized design. Herbages, consisted of dwarf napiergrass (*Pennisetum purpureum* cv. Mott) at 90% and Indigofera sp. at 10%, were fed at fresh matter basis 60%, with the experimental diet at 40%. Dry Matter Intake (DMI) tended to be higher in ED1 than in ED2 without significant difference (p>0.05) among treatments. The Average Daily Gain (ADG) decreased from 0.74 kg/head/day in ED0 to 0.43 kg/head/day in ED2, while that in ED1 did not decrease from ED0. The effect of experimental diets did not differ significantly (p>0.05) on body performance, hematological or biochemical parameters. The results indicated that the experimental diet did not affect the hematological or biochemical parameters; however, the feedstuff mixed with cocoa pulp at 5% showed a comparable average daily gain in fattening Bali steers with control.

Keywords: Biochemical and Hematological Property, Cocoa Pulp, Experimental Diet, Dry-Lot Fattening

Introduction

Feed cost represents about 70% in livestock production cost, the biggest component in livestock farming, which must prepare for feedstuff (Slattery, 2005; IPCC, 2014; Alqaisi *et al*., 2017. The price of feed fluctuates, depending on market demand and seasons such as the dry period. Moreover, pasture has been turned into settlements. Therefore, to meet the feed nutrition of livestock, farmers require not only additional feed sources for fattening cattle used to grow, produce and reproduce but also forage nutrition (Amsalu and Addisu, 2014).

Agro-industrial by-products and processed agro-industrial wastes are generally available as an alternative source of additional feed because they are quite abundant and cheaper than concentrates. Some researchers suggest that agro-industrial wastes from processed cacao such as cocoa pod husk and cocoa shell could supply the substantial energy requirements of ruminants, including cattle and sheep (Alexander *et al*., 2008; Alemawor *et al*., 2009; Adeyina *et al*., 2010). Additionally, one of the agro-industrial cocoa bean by-products potentially used as feedstuff is cocoa pulp, which is obtained from cacao processing (Rojo-Poveda *et al*., 2020). Some advantages of cocoa pulp are abundant production, potential nutritional value and feedstuff substitutability.

In Indonesia, especially South Sulawesi, cocoa is an export commodity. On the basis of the statistical data of South Sulawesi in 2019, one of the main commodities exported in 2019 was cocoa/chocolate, occupying 3.93% (SCSAS, 2018) and in fact, the Government of South Sulawesi is targeting cocoa production up to 276,000 tons/year. Hence, the availability of cocoa by-products (i.e., cocoa pulp) would possibly increase, representing sufficient feedstuffs in feed concentrates. Cocoa pulp is obtained when the beans are removed from the pod after harvest. The remaining parts of pod and hust of the cocoa represent between 2/3 and 3/4 of the total weight of the fruit, while they are usually discarded by local farmers after pod processing (Makinde *et al*., 2019).

Cocoa pulp contains a high sugar content, which can be used as a source of energy for livestock (Schwan and Wheals, 2004; Duarte *et al*., 2010). Fermented cocoa shell as feed has been used by cattle (Suryanto *et al*., 2017; Syapridus *et al*., 2016) and goat and sheep (Zain, 2009). Some research findings suggest that the cocoa pulp has also been used as a feedstuff in sheep concentrate at 15% (Ako *et al*., 2019) and has the potential to be used up to 10% incomplete feed with corn cobs as the fibre source for ruminants (Natsir *et al*., 2018). However, no studies were conducted on the utilization of cocoa pulp as dry-lot fattening Bali steers. The previously research was aimed to show the potential of cocoa pulp as a ruminant feed for both sheep and cattle.

Blood components are important for maintaining the whole body of livestock. The main function of blood is to deliver the oxygen needed by cells throughout the animal body. Blood also supplies body tissues with nutrients, transports metabolic wastes and contains several ingredients that protect the body from various diseases. Generally, the dietary pattern may be more useful to understand the influence of diets on the metabolic health of livestock. Furthermore, the relationship between dietary patterns and metabolite profiles, such as blood metabolite profiles is required to acquire livestock performance. Therefore, the objectives of this study were to evaluate the effect of mixed levels of cocoa pulp in the experimental diets on the performance and blood metabolite profiles of fattening Bali steers.

Materials and Methods

Experimental Die Formulation

Herbages as a main source of feed consisted of dwarf napier grass at 90% and *Indigofera* sp. at 10%, whereas feed concentrates as an experimental diet consisted of cocoa pulp, rice bran, coconut cake meal, shrimp waste meal, corn epidermis, milled corn, molasses and minerals. The ratio of herbages and experimental diets was fixed among treatments to be provided at 60 and 40%, respectively. The experimental diets were divided into three levels of cocoa pulp at 0, 5 and 10% as dry matter basis with substituting milled corn levels at 20, 14 and 8%, respectively, in ED0, ED1 and ED2. The feedstuff composition of the experimental diets is shown in Table 1. The chemical contents of experimental diets were analysed at Animal Product Technology, Faculty of Animal Science, Hasanuddin University, Makassar.

Composition of cocoa pulp consisted of 7.00% dry matter, 7.55% crude protein, 0.49% crude fat and 7.71% crude fibre and that of milled corn, 86% dry matter, 9% crude protein, 2% crude fibre and 4% crude fat. Furthermore, as the quality of the experimental diets, chemical properties and phytochemical components were analysed.

The chemical composition of the experimental diets was determined for the moisture content by oven-drying at 105°C based on the INS (1992) No. 01-2891-1992; the total Nitrogen (N) content, by the Kjeldahl procedure (AOAC, 1990); the percentage of crude protein, calculated as total N × 6.25; crude fibre, by INS (1992) No. 01-2891-1992; and ash, by heating at 600°C (AOAC, 1990). Flavonoid and theobromine composition of the experimental diets was determined using ultraviolet-visible spectrophotometry (Bojić *et al*., 2013) and the antioxidant composition, by the diphenyl-2-picrylhydrazyl method (Fitriana *et al*., 2016).

Site Description and Feed Trial

The experiment was conducted in Gowa Regency (South Sulawesi, Indonesia). Three experimental diets contained three replications, allocated into three heads and thus, nine heads of dry-lot fattening Bali steers were allocated in total. The experimental diets were fed once at 7:00 a.m. and herbages were fed twice a day at 10:00 a.m. and 4:00 p.m. The experimental diet was fed at 3% dry matter of body weight of steers. Drinking water was given *ad libitum*.

Performance of Dry-Lot Fattening Bali Steers

Performance of dry-lot fattening Bali steers, such as body weight gain (kg), chest circumference (cm), body length (cm) and shoulder height (cm), were taken after 2 weeks of feed adaptability as the initial day (D-0). At the end of the research, data were also collected as the final day (D-85).

Weight gain, chest circumference, body length and a shoulder height of fattening Bali steers were measured on the initial day, days 25, 55 and 85, designated as D-0, D-25, D-55 and D-85, respectively. Dry Matter Intake (DMI), Average Daily Gain (ADG) and Feed Conversion Rate (FCR) were calculated as shown in Eq. 1, 2 and 3, respectively.

$$DMI(kg/head/day) = (Total\ amount\ of\ feed\ consumed - Remaining\ feed)\ per\ day \quad (1)$$

$$ADG(kg/head/day) = \frac{Final\ body\ weight(kg) - Initial\ body\ weight(kg)}{Experimental\ period(day)} \quad (2)$$

$$FCR(kg/kg) = \frac{Amount\ of\ feed\ fed(kg)}{Experimental\ period(day)} \quad (3)$$

Blood Metabolite Profiles of Fattening Bali Steers

Blood samples of Bali steers were collected in EDTA tubes for each animal on D-0, D-25, D-55 and D-85 to determine hematological parameters such as Red Blood Cell (RBC) count, White Blood Cell (WBC) count, Hemoglobin (Hb), and Hematocrit (Ht). The percentages of RBC and WBC counts were determined by hemocytometry and Hb and Ht, by a hemoglobinometer and a microhematocrit centrifuge, respectively. Blood samples were analyzed at the Laboratory of Animal Physiology, Faculty of Animal Science, Hasanuddin University, Makassar, South Sulawesi, Indonesia.

Biochemical parameters of blood samples, such as glucose, cholesterol and blood urea, were determined by hexokinase, CHOD-PAP and enzymatic-GLDH, respectively, by the instruments from Thermo Scientific Indiko, Germany. Additionally, alanine Aminotransferase (ALT) and aspartate Aminotransferase (AST) were determined by International Federation of Clinical Chemistry, IFCC without pyridoxal phosphate (incubation at 37°C). The biochemical parameters were analysed at the Public Health Laboratory (Makassar, South Sulawesi, Indonesia).

Statistical Analysis

The data obtained were analysed using SPSS software for Windows ver. 16.0 (Chicago, IL, USA). The experiment was carried out according to a completely randomized design with three treatments and three replications and the difference among treatments was tested by the least significant difference at the 5% level.

Results

Chemical Analysis of the Experimental Diets

The chemical contents of proximate and phytochemical components in the experimental diets were measured (Table 2). As for the proximate components, the moisture content increased from 11.48% (ED0) to 42.88% (ED2) along with increasing cocoa pulp level from 0 to 10%, due to high moisture content (7% dry matter) in cocoa pulp. Similarly, crude protein, crude fibre and ash contents tended to increase, while fat and nitrogen-free extract contents tended to decrease, with increasing levels of the cocoa pulp as well as reducing levels of milled corn.

Effect of the Experimental Diets on Performance of Dry-Lot Fattening Bali Steers

Performance of dry-lot fattening Bali steers fed *in vivo* to three levels of experimental diets mentioned above were determined for DMI, ADG and FCR, which were tested by statistical analysis at 5% level (Table 3). With increasing levels of cocoa pulp as well as reducing levels of milled corn, DMI tended to decrease from 2.81% (ED0) to 2.37% (ED2) without significant difference among treatments, while ADG decreased ($P < 0.05$) from 0.74% (ED0) to 0.43% (ED2) and thus, FCR tended to increase from 4.53 (ED0) to 6.56 (ED2).

Performance of dry-lot fattening Bali steers with respect to body size parameters, such as weight gain, chest circumference, body length and shoulder height is presented in Fig. 1. The experimental diet did not significantly affect ($p>0.05$) neither on weight gain (Fig. 1A), chest circumference (Fig. 1B), body length (Fig. 1C), nor on shoulder height (Fig. 1D). In all the experimental diets, body weight increased from D-0 to D-85, while it showed stagnant on D-55 and turned to increase on D-85 in ED2 and tended to run through the higher values from D-25 to D-85 in ED1, compared with the other diets.

The chest circumference in ED1 tended to increase faster compared with that in ED0 and ED2 from D-25 to D-85. The body length at all experimental diets tended to be inconsistent over time; however, the body length in ED1 tended to be high on D-55. The shoulder height of ED1 tended to be higher than that of the other diets from D-25 to D-85.

Effect of the Experimental Diets on Blood Metabolite Parameters in Dry-Lot Fattening Bali Steers

The experimental diet did not significantly affect the profile of either the hematological or the biochemical parameters ($p>0.05$) (Table 4) and all the hematological and biochemical parameters entered to the normal threshold of cattle. As for the hematological parameters, RBC count tended to increase along with additional levels of cocoa pulp in the experimental diets. Although the high value of WBC can be used as a parameter of disturbed health of cattle, it did not affect the cattle performance of dry-lot fattening steers, or trends in ADG in the current study (Fig. 1A).

Although blood Hb and Ht values also fluctuated with the experimental diets, blood Hb and Ht values were still within the normal threshold. Hb in RBC carries oxygen throughout the body for metabolic processes and provides the colour of red blood cells and high level of Hb increases the oxygen supply to tissue; therefore, the metabolic processes in the animal tissues run optimally. The experimental diets affected different trends in biochemical properties, which did not differ significantly among diets, ranging from increasing trend in blood urea levels, decreasing trend in cholesterol and ALT levels and fluctuating trend in glucose and blood AST levels along with additional levels of cocoa pulp (Table 4).

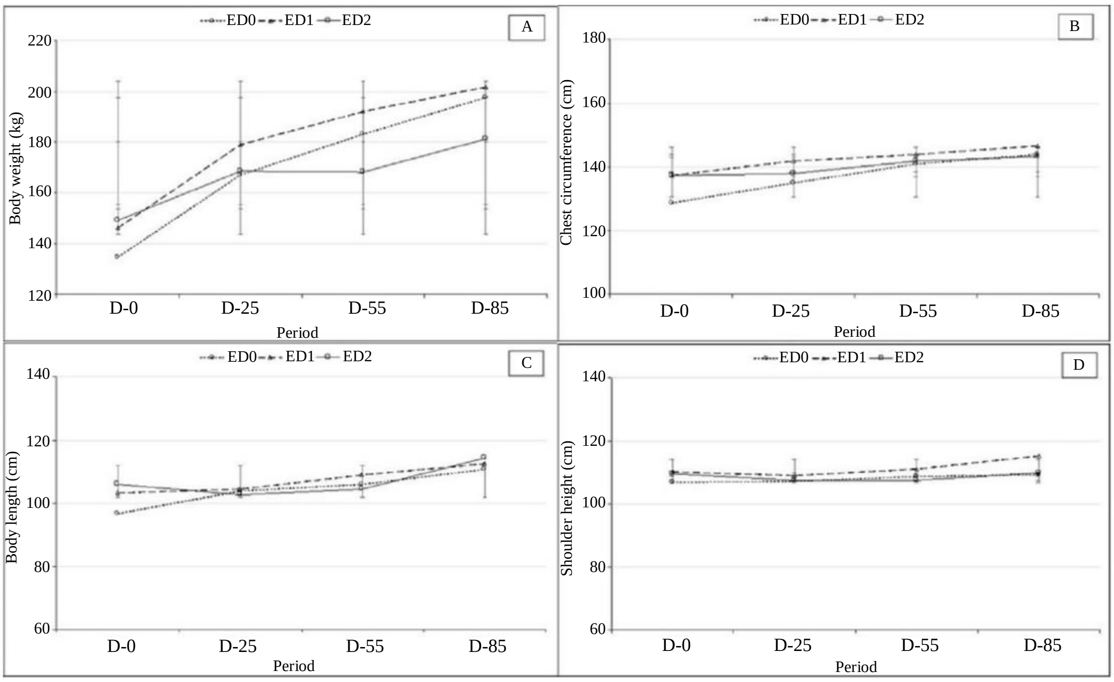

Fig. 1: Performance of dry-lot fattening Bali steers: (A) body weight gain; (B) chest circumference; (C) body length; (D) shoulder height. The experimental diets, ED0, ED1 and ED2 mean the feedstuffs mixed with 0, 5 and 10% levels of cocoa pulp, respectively

Table 1: The feedstuff composition of experimental diets for the performance of dry-lot fattening Bali steers

	Experimental diet (% cocoa pulp)		
Feedstuff (%) DM basis	ED0 (0%)	ED1 (5%)	ED2 (10%)
Cocoa pulp	0	5	10
Rice bran	20	20	20
Coconut cake meal	20	20	20
Shrimp waste meal	20	21	22
Corn epidermis	17	17	17
Milled corn	20	14	8
Molasses	2	2	2
Mineral	1	1	1
Total	100	100	100

Table 2: Chemical content of experimental diets for the performance of dry-lot fattening Bali steers

	Experimental diet (% cocoa pulp)		
Parameter	ED0 (0%)	ED1 (5%)	ED2 (10%)
Proximate component (%)			
Moisture content	11.48±0.33	33.43±0.86	42.88±2.56
Crude protein	12.43±0.64	12.88±0.44	13.76±0.24
Crude fat	5.37±0.43	3.79±0.46	3.89±0.20
Crude fiber	17.14±0.32	17.14±1.65	20.00±1.74
Ash	21.20±0.79	24.13±0.77	22.26±1.11
Nitrogen-free extract	43.86±1.16	42.06±1.12	40.09±1.09
Phytochemical component (%)			
Flavonoid	0.19±0.00	0.16±0.00	0.15±0.00
Theobromine	0.34±0.01	0.55±0.00	0.60±0.00
Antioxidant	77.09±7.69	47.68±4.29	47.11±7.55

Data were presented as means ± standard deviation. §ED0 = the experimental diets without cocoa pulp; ED1 = the experimental diets with 5% of cocoa pulp; and ED2 = the experimental diets with 10% of cocoa pulp. Nitrogen Free Extract = 100% DM – Crude Protein – Crude Fat – Crude Fiber – Ash

Table 3: Dry matter intake, daily body weight gain and feed consumption rate of dry-lot fattening Bali steer

Parameter	Experimental diet (% cocoa pulp) ED0 (0%)	ED1 (5%)	ED2 (10%)
DMI (kg/head/day)	2.81±0.09	2.77±0.92	2.37±0.44
ADG (kg/head/day)	0.74 ± 0.10^{a}	0.70 ± 0.14^{ab}	0.43 ± 0.21^{b}
FCR	4.53±2.02	5.84±3.10	6.56±2.45

Data were presented as means ± standard deviation. Values with superscripts in the same row is differ significantly (P<0,05). ns = not significant (P>0.05).; DMI = Dry matter intake; DBWG = Daily body weight gain; FCR = Feed Consumption Rate. Experimental diet refers to Table 1

Table 4: Blood metabolite parameters of dry-lot fattening Bali steers

Parameter	Experimental diet (% cocoa pulp) ED0 (0%)	ED1 (5%)	ED2 (10%)
Hematological†			
RBC count (10^6/mm^3)	4.81±7.86	5.34±3.51	5.37±3.58
WBC count (10^3/mm^3)	9.43±0.95	9.78±1.94	9.60±1.10
Hb (g/dL)	8.60±0.92	8.40±0.92	9.60±1.35
Ht (%)	28.00±2.00	31.67±2.52	32.00±4.58
Biochemical‡			
Glucose (mg/dL)	42.33±2.52	42.67±2.31	38.00±7.00
Cholesterol (mg/dL)	161.00±9.17	148.00±14.42	126.67±45.45
Blood urea (mg/dL)	22.33±2.08	22.00±3.00	28.00±4.00
ALT (U/L)	59.33±8.62	55.33±6.66	45.67±3.21
AST (U/L)	80.67±7.63	74.67±7.09	81.00±6.93

†Laboratory of Animal Physiology, Faculty of Animal Science, University of Hasanuddin, Makassar, South Sulawesi, Indonesia, 2018. ‡Public Health Laboratory, Makassar, South Sulawesi, Indonesia, 2018. RBC = Red Blood Cell; WBC = White Blood Cell; Hb = Haemoglobin; and Ht = Haematocrit. ALT = Alanine Aminotransferase; AST = Aspartate Aminotransferase. Experimental diet refers to Table 1

Discussion

Crude protein contents ranging in 12.4-13.8% in the present diets are adequate for optimal microbial protein synthesis (Hume *et al*., 1970), although if this level drops below 7%, the rumen microorganisms cannot break down the feed efficiently and addition protein supplements need to be added. The whole experimental diets in ED0 to ED2 sufficiently meet the nutritional needs of cattle according to the Indonesian National Standard 2017 No. 3148-2. A nutritionally balanced diet, including sufficient nitrogen and rumen degradable protein must be provided to maximize bacterial fermentation, feed intake and energy digestibility in feeding to ruminants (Matthews *et al*., 2019).

As for the phytochemical components, the flavonoid and antioxidant contents decreased with adding cocoa pulp, while the mixed level of cocoa pulp in a range of 5-10% affected these two phytochemical components negligibly. Flavonoids are used to be correlated with their antioxidant activities pharmacologically (Gryglewski *et al*., 1987). Providing 5% level cocoa pulp (ED1) to experimental diets containing flavonoids and antioxidants is expected to improve the meat quality, especially related to health issues. According to Heim *et al*. (2002), flavonoids are polyphenol compounds that are antioxidants or free radical scavengers, have anti-inflammation properties and prevent DNA damage in animal cells (Table 2).

In contrast, the theobromine content increased along with additional levels of cocoa pulp to reach the highest level in ED2 (Table 2). Theobromine content can affect the following disorders: Abnormal spermatozoa morphology, emptying of Sertoli cells, imperfect fetal growth, weight loss, miscarriage and animal death. Furthermore, providing cocoa pods in sheep and goat feed containing theobromine antinutrients can reduce the consumption of dry matter live weight gain (Alexander *et al*., 2008).

Neither DMI nor ADG did not decrease from ED0 to ED1, while decreased substantially from ED1 to ED2, possibly due to the highest percentage of theobromine in the diet, ED2, since (Alexander *et al*. 2008) argued that the high percentage of theobromine in the feed formulation could inhibit the growth of rumen microbes and decrease the ability to digest fiber. Therefore, 20% crude fibre level in ED2 may lead to intricate rumen-microbe digestion, According to Mahyuddin (1995) live weight gain began to decline at 37% of cocoa shell (or 15% of the ration). At this concentration, the ration contains 0.24 percent theobromine, may contribute to lower metabolizable energy consumption. Drop of DMI in ED2 may be related with the highest fibre content among treatments, since the increase in crude fibre results in lowering animal productivity.

According to (Ako *et al* 2019), the use of 10% cocoa pulp in the complete feed formulation containing different sources of herbages, either rice straws, corn cobs or soybean straws, promotes the performance of local goats. Although the experimental diet did not significantly affect DMI, DMI is usually used as an indicator of the level of diet palatability. The provision of exceeding 5% level of cocoa pulp as feedstuff in the feed concentrate could decrease the palatability of concentrate feed.

Similar to DMI, ADG decreased substantially from ED1 to ED2, while the decrease in ADG from ED0 to ED1 was negligible (Table 3). The current ADG levels in ED0 and ED1 were still higher, compared with the previous study on male Bali cattle, where the cattle were fattened with *ad libitum* field grass and 1% concentrates to reach 0.69-0.72 kg/day ADG.

FCR is feeding conversion to determine the amount of feed converted into body weight gain and is used as an indicator in feed efficiency. As stated by (McCrickerd and Forde, 2016), FCR affected by palatability depends on several factors such as feed appearance, feed shape, door, taste and texture of the diet. In the current study, FCR increased steadily from 4.53, 5.84 to 6.56 along with additional levels of cocoa pulp from 0, 5 to 10%, respectively, which was more efficient in FCR compared with 7.32 in the previous study (Melati *et al*., 2019). Increase in FCR means the increase in feeding cost paid by farmers and animal weight gain is inversely proportional to the FCR value. Therefore, cocoa pulp feeding to fattening Bali steers could be provided within 5% level.

The animal performance of dry-lot fattening Bali steers in ED1 tended to give the best results compared with ED0 and ED2, which was derived from the following mechanisms: The chemical composition of the experimental diets in ED1 being better, showing higher palatability associated with DMI and the higher ADG (Table 3). The experimental diets in the current study followed the fattening cattle feed standard enacted by the Indonesian National Standard. Additionally, consumption is an important factor in determining ruminant productivity and animal body size (Aregheore, 2000). Chaokaur *et al.* (2015) stated that the higher the level of feeding, the higher the daily gain of Brahman cattle. Body weight gain generally increases due to the capacity of the digestive tract (Heryiani *et al*., 2020), unless the feeding level is limited. Food nutrients are converted by the digestive system into simpler chemicals that can be absorbed and used as energy (Heryani *et al*., 2020) However, a shorter digesta retention period in smaller herbivores may limit the nutrients availability (Ramzinski and Weckerly, 2007)

Increased levels of RBC count indicate that the oxygen content in the blood may increase, which has a positive impact on the body's metabolic activity. The RBC count in cattle is ranging in 3.8-5.7 million/mm^3 (Utama, 2001), 5.49 million/mm^3 (Adam *et al*., 2015) and 6.33-8.89 million/mm^3, which tends to be higher than in the current study where the RBC count was still within the normal threshold. The WBC count of fattening Bali steers maintained within 9.43-9.78 thousand/mm^3 (Table 4).

The blood glucose level found highest in ED1 entered lower value than the previous report (Tahuk *et al*., 2018), while higher than the other (Kamal *et al*. 2016). Animal feed is one of the factors that affect the level of blood glucose content, since (Tahuk *et al*., 2018) stated that feed for energy consumption is very important to determine the level of blood glucose. Cocoa pulp, containing glucose at 214.24 g/L, fructose at 10.6 g/L and sucrose at 107.6 g/L, has the potential for use as a feedstock for diet energy (Gyedu, 2001). Although the glucose levels in the current study were lower than the previous studies, they were still within the normal threshold of 43-100 mg/dL (Mitruka and Rawnsley, 1977. Furthermore, the presence of flavonoid content as an antioxidant in cocoa pulp maintains blood glucose stability, stating (Akhlaghi and Bandy, 2009) that the antioxidant activity of flavonoid compounds can capture free radicals to protect pancreatic β-cells. This protection mechanism plays an important role in preventing damage and consistently maintaining insulin production in the animal body.

The blood cholesterol levels tended to decrease along with the additional levels of cocoa pulp, showing highest cholesterol level found in ED0 (Table 4), in which the level was similar to the previous study (Suharti *et al*., 2009), who stated that the range of cholesterol in cattle (i.e., 159.75-193.25 mg/dL) was lower than in dairy cows (Faza *et al*., 2017) (i.e., 221.18-250.16 mg/dL). The value of blood cholesterol in the current study was still within the normal threshold value of 80-180 mg/dL. The high cholesterol levels in ED0 may be due to the highest crude fat level, compared with ED1 and ED2 (Table 2), since the feed fat level can increase the total cholesterol levels in the blood (Sejrsen *et al*., 2006). However, the present experimental diets did not result in changes in fat metabolism in the rumen. The blood cholesterol in the experimental diets containing cocoa pulp results in reducing the content, compared with control.

The blood urea levels tended to increase from ED1 to ED2. The levels of urea in ED0 and ED1 were lower than that of the previous study, showing 24.11 mg/dL (Tahuk *et al*., 2018). Blood urea is used as an indicator to assess the adequacy of animal feed protein during the fattening period. If the ammonia level in the rumen is high, the absorption of ammonia carried to the liver will be excessive. The enhanced concentration of blood protein urea will be proportional to the availability of feed protein (Getahun *et al*., 2019).

The blood ALT levels tended to decrease along with additional levels of cocoa pulp, showing that the experimental diets containing cocoa pulp reduced blood ALT levels, compared with the control (ED0). This is probably due to the inadaptability of the experimental diets and additionally, the examined steers experienced changes in feeding patterns, triggering a level of stress on them. Xie *et al.* (2020) stated that the liver and kidney tissues can experience disturbed metabolism if there is a change in feeding patterns. Furthermore, Picard *et al.* (2018) mentioned that apart from feeding patterns, stress can affect the metabolic processes of livestock, where the mitochondria work excessively to produce excess energy, resulting in cell damage in blood. Table 4 shows that the value of ALT levels was 45.67-59.33 U/L, which entered still normal range, according to the previous study, ranging around 20-76.8 U/L (Mitruka *et al*., 1997).

AST can be used as an indicator of the health of liver function. The liver is the center of carbohydrate, protein, fat and mineral metabolisms, all of these related to feedstuff. Feed contaminated with hazardous materials will generate metabolic damage in the liver (Ali *et al*., 2019). On the basis in Table 4, AST values in this study fluctuated in the range of 58-83 U/L, which entered still within the normal threshold for cattle, according to the previous study, finding AST value around 8.5-93 U/L (Mitruka *et al*., 1997).

The present experimental diets provided in fattening steers meet the Indonesian National Standard and did not jeopardize either the hematologic or biochemical functions of fattening Bali steers. Therefore, the present experimental diet was suitable for consumption of the steers.

Conclusion

The results suggest that the experimental diets mixed with 5 and 10% levels of cocoa pulp did not affect the hematologic or biochemical parameters of fattening Bali steers. However, the mixed level of cocoa pulp with concentrates should be recommended at the level of 5%, due to no decrease in average daily gain or dry matter intake from control. In the future, feeding 5% cocoa pulp to dry-lot fattening Bali steers could be implemented with reducing rate of milled corn and reducing wastes as using by products from the cocoa industry.

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Declaration of Interest Statement

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

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Author's Contributions

Renny Fatmyah Utamy and Ambo Ako: Conceived and designed the experiments, performed the field experiments, performed analyzed data, wrote the paper

Yasuyuki Ishii: Experiments performed analyzed data, wrote the paper

Muhammad Ihsan Andi Dagong: Performed the field experiments, performed analyzed data, wrote the paper.

N. Nahariah: Performed phytochemical analysis.

Purnama Isti Khaerani, Arung Bandong and Ardianto: Performed the field experiments.

Faisal Asbar: Performed the field experiments, performed hematology analyzsis

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues involved.

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