Estimation of microbial protein supply on goat fed on complete feed containing cocoa pulp with different fiber sources

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Abstract: Supplementation both energy and protein of goats can support optimal microbial growth so that utilization of fibrous feed can be optimized. The aim of this research is to observe the effect of goats which fed complete feed containing 10% cocoa pulp and various fiber sources toward estimation of microbial protein supply (MPS) and efficiency of microbial protein supply (EMPS). Twelve local male goats, aged 1-1.5 years old, were randomly assigned to one of four treatments according to completely randomized block design consisted of 3 treatments and 4 blocks as replications. The treatments were P1: Complete feed containing 50% rice straw, P2: Complete feed containing 50% corn cobs and P3: Complete feed containing 50% soybean straw. The results showed the estimated of MPS was P1 = 16.25 g/d P2 = 14.05 g/d, and P3 = 24.57 g/d, respectively. The EMPS per kg organic matter fermented in rumen was P1 = 86.5 g/kg BOFR, P2 = 73.0 g/kg BOFR, and P3 = 150.25 g/kg BOFR. Analysis of variances indicated that treatment did not affect (P>0.05) MPS and EMPS. In conclusion, based on estimated MPS and EMPS, the three types of fiber could be used as the main fiber sources in formulating complete feed containing cocoa pulp for local goats.

1. Introduction
Cocoa (Theobroma cacao, L.) is one of the export commodities that has a strong position in the world market. Indonesia is among the top 6 cocoa producing countries in the world. Cocoa production reaches 2.2 % of world cocoa production[1]. South Sulawesi, one of the provinces in Indonesia, has the largest cocoa production which reaches 494,241 tons[2]. Fermented cocoa beans will produce waste in the form of a cocoa liquid which amounts to 5 liters/ton of cocoa. Generally, the waste is disposed into the river without any treatment thus pollute the river[3]. Other studies report that the waste of cocoa pulp reaches 100-190 ml from 1 kg processed cocoa[4]. Products from cocoa pulp are herbicides for plants[5, 6], material for various food products and beverages such as jams, juices, and fermented drinks[7, 8]. However, production of cocoa pulp is high because the utilization of the material is very limited so that the cocoa pulp is very potent as a waste that can pollute the environment. An alternative to utilizing cocoa pulp is used as an additional feed ingredient in complete feed for ruminants. The cocoa pulp contains lots of glucose which can be used as a source of energy for livestock. There is approximately 25-30% of seed weight which contains sugar with relatively high concentration. The sugar has approximately 10-13% which have the potential as energy sources of feed material[9]. Cocoa pulp also has the potential to be used as molasses substitutes in complete feed...
ingredients. The complete feed can be prepared from a mixture of agro-industrial waste and agricultural waste which has not been used optimally so that the livestock does not need to be forage-fed[10]. Several studies have reported that providing complete feed by using agricultural waste such as corn cobs can improve the performance of local goats in South Sulawesi [11], can increase consumption and digestibility in male pepper goats [12], and can improve the quality and production of goat milk [13]. However, application of cocoa pulp in complete feed can only up to 10%[14]. Microbial protein has an important role for ruminants. Microbial protein contributes 70-100 percent of availability of the protein total for livestock. Measurement of microbial proteins of the gastrointestinal tract is very important to estimate ruminants protein needs [15]. One of the methods to predict the microbial protein synthesis is the measurement of purine derivatives which are contained in urine. Purine derivatives are mainly from microbes [16]. The excretion of purine derivatives can be used as an indicator of rumen microbial protein supply for the animal. The previous description is the background of the research on estimating microbial protein intake of goats fed complete feed containing cocoa pulp with different fiber sources.

2. Materials and methods

2.1. Formulation of complete feed

Formulation of the complete feed on the research is as the following protocols: All feed ingredients having large or rough particle size, such as corn cobs, rice straw, soybean straw were grinded to be smaller using milling machine. Each feed ingredient was weighted according to the formula composition (three different treatments) then all feed material was mixed by using a hand mixer. The feed ingredients composition of each treatment is presented in table 1.

2.2 Complete feeding for goats

This study used 12 male goats with the age 1 year with a body weight of 10-20 kg. The goats are placed in a metabolic cage with food and water. Feed is given twice a day at 08.00 and 16.00 with the same portion. Water is given by ad libitum.

2.3 Data sampling

Data sampling was carried on during the last 5 days of each period. The collected data is included leftovers and feces. Each leftover and feces were collected then it was weighted for 5 days of the sampling period. After that, the samples mixed homogeneously then picked out as much as 10% for laboratory analysis.

2.4 Laboratory analysis and calculation

The observed variables in this study were excretion of purine derivatives in urine and efficiency estimation of microbial protein intake.

Digested Organic Material (BOT) (g/day):

\[ \text{BOT} = \text{DCBO} \times \text{BO consumption} \]

The concentration of Purine Derivative (mmol/L) is the total purine derivative in urine which is the total of allantoin, uric acid, xanthine, and hypoxanthine.

Estimation of Microbial Protein Supply (g/day) is the amount of absorbed microbial purine derivatives (PD) (X, mmol/ day) related to excretion of purine derivatives (PD) in urine (Y, mmol/ day) which calculated based on the equation[16]:

\[ Y = 0.84X + (0.150W^{0.75}e^{-0.25x}) \]

Estimation of microbial nitrogen supply (NM) (g N / day) is calculated based on Chen dan Gomez recommendation (1992)[16]:

\[ \text{NM} = \frac{70X}{0.116 \times 0.08 \times 1000} = 0.727X \]
Efficiency estimation of microbial proteins based on the fermented digestive organic material on the rumen (g/ kg BOTR) which is calculated using the formula:

\[
\text{Efficiency Estimation of N Microbial Proteins} = \frac{\text{Estimated N in Feed}}{\text{Estimated N in Rumen}} \times 100\%
\]

The percentage of efficiency estimation of microbial protein based on N intake (%) which was calculated using the formula:

\[
\% \text{ Estimation of N Microbial} = \frac{\text{Estimated N in Feed}}{\text{N intake}} \times 100\%
\]

### Table 1. Complete Feed composition according to the treatment

| Feed material (%) (DM basis) | Treatments |
|-----------------------------|------------|
|                             | P1         | P2         | P3         |
| Rice straw                  | 50         | 0          | 0          |
| Corn cobs                   | 0          | 50         | 0          |
| Soybean straw               | 0          | 0          | 50         |
| Rice bran                   | 20         | 20         | 20         |
| Coconut cake meal           | 9          | 9          | 9          |
| Cocoa pulp                  | 10         | 10         | 10         |
| Shrimp waste meal           | 6          | 6          | 6          |
| Salt                        | 2          | 2          | 2          |
| Mineral Mix                 | 2          | 2          | 2          |
| Urea                        | 1          | 1          | 1          |
| **Total**                   | **100**    | **100**    | **100**    |

P1 = Complete feed containing rice straw 50%, P2 = Complete feed containing corn cobs 50%, P3 = Complete feed containing soybean straw 50%.

### 2.5 Data analysis

Data were analyzed by analysis of variance according to 3 x 4 randomized block design (RBD), 3 treatments and 4 groups. If the treatment has a significant effect, further testing was carried out using Duncan's Multiple Range Test [17]. Data were analyzed using the SPSS 16.0 statistical program with a mathematical model

\[
Y_{ijk} = \mu + \tau_i + \beta_j + \epsilon_{ij}
\]

### 3. Results and discussion

Based on analysis of chemical composition of complete feed with various fiber (P1=complete feed with 50% rice straw; P2=Complete feed with 50% corn cobs; P3=Complete feed with 50% soybean straw), the result can be seen in table 2.

The crude protein in the complete feed of P1, P2, P3 is 15.38%, 12.69%, 16.83%, respectively. The results of the analysis show that the crude protein in each treatment was able to meet the needs of goats. Dietary feed that is usually given to goats and sheep livestock ranging from 9-12%[18]. With the range of crude protein, it is also can meet the protein needs of livestock and help the development of rumen microbes. Rumen microbes will be able to be well developed when the amount of crude protein on dietary feed is 13.4%[19].
Table 2. Chemical Composition Of the Complete Feeda

| Components (g/kg/DM) | P1   | P2   | P3   |
|----------------------|------|------|------|
| Dry material         | 88.69| 85.64| 77.67|
| Crude protein        | 15.38| 12.69| 16.83|
| Crude fat            | 6.36 | 2.76 | 5.77 |
| Crude fiber          | 19.04| 29.30| 28.53|
| Ash                  | 11.31| 14.36| 22.33|
| ADF                  | 30.27| 37.96| 27.98|
| NDF                  | 43.37| 53.64| 42.19|
| Cellulose            | 23.79| 22.35| 18.93|
| Hemicellulose        | 13.10| 15.68| 14.21|
| Lignin               | 6.44 | 15.59| 9.03 |
| ATL                  | 0.04 | 0.02 | 0.02 |

*Animal Feed Chemical Laboratory, Hasanudin University, 2018. P1 = complete feed with rice straw 5%, P2 = complete feed with corn cobs 50%, and P3 = complete feed with soybean straw 50%.

Table 3. Total Excretion of Purine Derivatives and Estimation of Microbial Protein Intake

| Parameters                                         | Treatments |
|---------------------------------------------------|------------|
|                                                   | P1   | P2   | P3   |
| Organic Material Consumption (g/day)              | 380  | 386  | 380  |
| Digested Organic Material (g/ day)                | 298  | 296.53| 284.64|
| Fermented Organic Material in the Rumen (g/ day)  | 193.70| 192.75| 185.02|
| Purine Derivative Excretion (mmol/ L)              | 1.28 | 1.07 | 1.73 |
| Estimation of Microbial Protein Intake            |       |      |      |
| N Microbial (g/ day)                              | 16.25| 14.05| 24.57|
| Estimation of Synthesis Efficiency of N Microbes  |       |      |      |
| N Microbes (g/ kg BOFR)                           | 86.5 | 73   | 150.25|
| N Microbes (%)                                    | 0.32 | 0.32 | 0.41 |

P1=Complete feed with rice straw 50%, P2=Complete feed with corn cobs 50%, P3=Complete feed with soybean straw 50%. Superscript non significants ( P>0.05).

The results of variance analysis show that the treatment of complete feed containing cocoa pulp with various fiber sources is non significants (P>0.05) on the consumption of organic material, digestible organic material, fermented organic material in the rumen, purine derivative excretion, estimation of microbial protein intake and efficiency estimation of microbial protein intake in goats. Consumption of organic material ranges from 380-386 g/day with an average of 382 g/day. Organic material consumption is affected by feed consumption which one of that is level of preference and taste of the feed were one that influences feed consumption is the level of preference and taste of the feed ingredients given. The amount of feed consumption is affected by palatability[20]. Palatability depends on the smell, taste, and appearance of the feed. The sufficient protein and finer in the structure size of feed can increase the amount of food consumption[21]. On the complete feeds, the cocoa pulp which was mixed with fiber sources such as rice straw, corn cobs, and soybean straw cannot be
separated from other materials such as coconut cake, bran, and etc because they are homogenous so that the livestock cannot choose feed ingredients.

Digestible organic material (BOT) describes the amount of organic material that digested in the rumen from consumed organic material. Obtained BOT values ranged from 284.64 g/day - 298 g/day. The BOFR value ranges from 185.02 - 193.70 g/day. The average of BOFR is varied according to the treatment in line with the BOT value. This is because BOFR is a BOT multiplication with 0.65[22]. A higher BOFR value was obtained from the previous study by giving a corncob-based complete feed with various protein sources ranging from 202.22 - 255.26 (g/day)[23]. Trend of digested organic material will depend on the trend of organic material consumption and the digestibility of organic material so that the higher of consumption and digestibility, the higher of digestibility organic material value[12]. The value of the fermented organic material in the rumen is also related to the value of the digested organic material.

The total excretion of purine derivatives of P1, P2, P3 is obtained 1.28 mmol/ L, 1.07 mmol/ L, and 1.73 mmol/ L, respectively. The value total of obtained purine derivatives is lower than previous studies. The value total is 6.08 mmol/L which is compared with 7.67 mmol/L. The supplementation of previous studies are oat hay and urea-enriched faba or barley[24]. The concentration of purine derivatives is affected by several factors, such as the level of consumption and weight of cattle [16, 25]. The excretion of purine derivatives can be used as an indicator of rumen microbial protein supply of the animal. Purines in the digestive tract that enter the small intestine are mostly from microbes. Purine derivatives which are excreted in the urine are mainly from microbes [16]. The purines are metabolized by the body of the ruminants and then removed together with urine in the form of purine derivatives. Purine derivatives are allantoin, uric acid, xanthin and hypoxanthin [26].

The concentration of purine derivatives which are excreted in urine is used to estimate microbial protein supply. The highest value of N microbes supply estimation is 24.57 g/day and the lowest one is 14.05 g/day. The amount of N microbes supply estimation is determined by the number of microbes that are absorbed into the intestine and become a source of protein for the host. The obtained value of N microbes supply estimation is higher than previous studies that it was fed by complete feed using various protein source ranging from 2.52-4.07 g/day[27].

The average of synthesis efficiency of microbial protein ranges from 73-150.25 g/kg BOFR. Generally, wide variations of microbial protein synthesis estimation have been reported in various studies and the results are still within the range of values which is from 14-49 g/ kg BOFR[22,16]. There are many things that affect the value of the estimation number, such as the difference in the method of estimation [28].

4. Conclusion
Based on estimated MPS and EMPS, the three types of fiber could be used as the main fiber sources in formulating complete feed containing cocoa pulp for local goats.

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