

Effect of Fermented Cocoa Pod Husk Level in Concentrate on Etawah Grade Semen Quality

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Abstract

The purpose of study was to determine the effect of fermented CPH supplementation level in concentrate toward semen quality of Etawah Grade Buck (EGB). This research was conducted from August to October, 2011 at experiment pen and Feeds Science Laboratory, Faculty of Animal Science, Universitas Gadjah, Yogyakarta. Semen was collected from 4 heads of EGB with about 2 years old and weight approximately of 24 kg. Concentrate was main material of CPH fermentation with composition P0 0, P1 20%, P2 40% and P3 60% from total of ration. The composer was used by the *Aspergillus niger* BPT with density 9.1×10^7 of colony/g as many as 1% of CPH material (on dry matter). Semen collection was done every 1 time/week and to be done five times semen collection. An examination was consisting of volume, color, smell, consistency, and the pH. Meanwhile, for microscopic examination was motion of mass, individual, motility, viability of spermatozoa. pH of P0, P1, P2 and P3 were 6.95, 7.03, 7.50 and 7.79, respectively. Mass movement of P0, P1, P2 and P3 were +++, +++, ++ and ++, while viability of P0, P1, P2 and P3 were 79.57%, 79.47%, 59.97% and 51.13%, respectively. It was concluded that addition of CPH fermentation in concentrates up to 20% for of EGB has not affect to feed consumption but it has affect to decreasing of semen quality in both of microscopic and microscopic.

1. Introduction

The low birth rate goat is usually caused malnutrition especially on ranch system. The malnutrition pregnant goat is giving birth to kid be weak and milk less available. This condition is generally causing death for kid. One of the objectives of this program was acceleration of increases in population through improving the reproductive system of goat. The goat farming is expected to make contributions of meat self-sufficient program because goat population in Indonesia is quite high. National data for goat population in Indonesia on 2016 about 19.608.181 heads, which it is the highest population in Central Java reached 4,140,130 heads and followed by East Java about 3,267,954 heads. While the population highest in the Kalimantan Island is North Kalimantan reached 160,230 heads and followed by East Kalimantan about 63,114 heads [1].

Increasing in population of the goat is through improving the reproductive system particularly the semen quality of semen. The good quality of semen has high fertilization when he has mating to ewe with high of pregnancy. One of the factors can affect on the goat semen quality with giving feed quality. The preparation of feed quality can use an agricultural waste that it is abundant available



during harvest season. The cocoa pod husk (CPH) is an alternative as main matter for ruminants concentrate. The CPH is always available on farmers cocoa plantation which it can be harvested throughout the year [2].

The benefits value of CPH as a fodder can be increased by reducing of fraction fiber like hemicellulose, cellulose and lignin [3]. CPH has low crude protein content but it has high crude fiber content about 9.14 and 35.74%, respectively, while content of fraction fibers like neutral detergent fiber (NDF) and acid detergent fiber (ADF) are 58.78% and 47.04%, respectively [4], hemicellulose, cellulose and lignin are 6.01%, 35.33% and 38.78%, respectively [5].

The decline of CPH fiber content fraction is fermentation methods using microorganism in both of aerobic or anaerobic conditions. Feeding of concentrates to Etawah Grade Buck (EGB) can improve the semen quality. The semen quality is determined by the ejaculate volume, color, pH, consistency, viability, motility abnormality and concentration of spermatozoa [6]. The purpose of study was to determine the effect of fermented CPH supplementation level in concentrate toward semen quality of Etawah Grade Buck (EGB).

2. Materials and Methods

This research was conducted at pen experiment and Feeds Science Laboratory, The Faculty of Animal Science, Universitas Gadjah, Yogyakarta. Equipments were used in this research such as artificial vagina (minitub), microscope, object glasses, glass cover, test tube, pipet, neubauer haemocytometer improved, tissue paper, universal of pH indicator (merck), hand counter, refrigerator, ice container, thermometer, hummer mill. Materials were used EGB semen, sterile water warm, vaseline, eosin, physiological NaCl 0.3 %, alcohol 70 %, feed, vitamins and drugs.

Semen was collected from 4 heads of EGB with about 2 years old and weight approximately of 24 kg. The health reproductive condition of animals should be normal, before feeding trials were given worm drug. Concentrates were given from the CPH with a measure that was enumerated 1x5 cm, which it was fermented in aerobic condition. The composer was used by the *Aspergillus niger* BPT with density 9.1×10^7 of colony/g as many as 1% of CPH material (on dry matter). Concentrate was main material of Fermentation CPH with composition 0, 20% (124 g), 40% (287 g) and 60% 482 g) from total ration (Table 1).

The CPH was fermented in seven days. Drying of CPH after fermentation process was 3-5 days depend on irradiating the sun. CPH dry was grounded using hummer mill by the size of screen in 5 mm. Feed was given to animals in form of fresh forage for King grass (*Pennisetum hybrid*) and concentrate which it was the portion as follows:

Table 1. The Portion Feeding Trials

Feed materials	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
King grass				
(<i>Pennisetum hybrid</i>) (g)	3,350	2,605	1,860	1,116
Groats soybean (g)	90	26	13	6
Fermented CPH (g)	-	124	297	482

The total of concentrates protein in concentrate was 12%. Feed was given every day to animals of 4.0% from live weight on dry matter basis [7]. Concentrates were enriched by the addition of 2.0% minerals and 0.5% salt from total of concentrates material. Feeding trials were done twice a day, morning at 08.00 AM and 03.00 pm, started feeding of concentrates and to be continued by King

grass chopping. Adaptation period of feed trial was done 15 days before started of the research. Water drinking was given unlimited (*ad-libitum*).

The semen collection was done using artificial vagina already cleaned, sterile and dried. Warm water with the temperature 45° C incorporated into the artificial vagina, then in the interior of the vagina artificial mouth was lubricated by vaseline. Teaser prepared was used to increasing the libido and get the good quality and to be done false mounting [8]. Semen collection was done every 1 time/week and to be done five times semen collection. Semen collection was done started at 09.00 AM after feeding. The frequency of collection was three times for each individual treatment. Semen was collected to be sheltered in tube inside and put it in a ice container and taken to laboratory for next analysis. Variable was observed in microscopic examination. An examination was consisting of volume, color, smell, consistency, and the pH. Meanwhile, for microscopic examination was motion of masses, individual, motility, viability of spermatozoa according to procedures of [9].

Motility of spermatozoa or impulse spermatozoa was determined in the mass movement and the movement of individuals. Based on the results of the assessment of the mass movement of semen quality can be determined as follows: a) excellent (+++), which it is looks great waves, thick, dark, and many, like wisps of active black clouds move fast moving place; b) good (++), when visible waves of small, thin, sparse, less clear, and moving slowly; c) fair (+), if not visible waves but rather just individual movements active progressive; d) (N, necropermia or 0) if only little or no movement of the individual.

Semen concentration was the number of spermatozoa per ml of semen. Directly calculation with hemocytometer that was by using a method of neubauer calculating room. The concentration of spermatozoa in the five chambered calculated according to a diagonal direction using a microscope with an enlargement 40x10. The formula to calculate the concentration of spermatozoa:

$$Y \times \frac{400}{80} \times \frac{200}{0.1} = Y \times 10,000 \text{ mm}^3 = Y \times 10,000,000/\text{ml}$$

Y = Totally of observation spermatozoa in 5 big box
 400 = Totally of small box in calculation room
 80 = Totally of small box in 5 big box
 200 = Dilution of 200 times
 0.1 = Volume of calculation box (mm³)

The percentage of life spermatozoa was a drop of a substance with eosin in one glass object sterile and one small drops sperm added and mixed evenly, to be observed under a microscope with 40x10 enlargement. Calculation of 100 to 200 semen cell and the best 500 semen cells [8] using hand counter. The life spermatozoa head was not absorb color but it was dead absorb a red color after that the percentage of the life spermatozoa to be determined.

The percentage of spermatozoa abnormally was after counting of spermatozoa concentration and this semen was used to calculate the percentage semen abnormally. Spermatozoa was calculated up to 200 cells on one or several views under the microscope 40x10 with enlargement, then to be determined the percentage of spermatozoa abnormally.

3. Results and Discussion

3.1. Feed Consumption

The total of feed consumption in the first day and eighth day of the feeding trial is presented in Table 2.

Table 2. Feed consumption

	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Feed consumed* (daily):				
King grass (g)	2,727.47	2,366.45	1,654.90	1,116.00
Groats soybean (g)	90.00	26.00	13.00	6.00
Fermented CPH (g)	-	61.90	50.38	141.01
Feed consumed** (daily):				
King grass (g)				
Groats soybean (g)	3,069.66	2,538.79	1,825.91	1,116.00
Fermented CPH (g)	90.00	26.00	13.00	6.00
	-	80.15	197.71	298.39

* Feed collected in first day of feeding trial.

**Feed collected in eighth day of feeding trial.

Feeding trial of P₀ as a comparison that is not granted an additional CPH fermentation in concentrate so that the portion of the King grass more as basal feed and fiber source. While the P₁, P₂ and P₃ a bit more given the grass King because given additive of CPH fermentation as fiber source. The addition of groats soybeans were all treatments as the main protein source in the concentrate. According to [10, 11] groats soybeans contain crude protein reached 41.3-41.7%. The addition of groats soybeans in rations on this research can fulfill the standards of the crude protein needs of animal growing about 12%.

The high feed consumption on P₀ was caused by feeding only from King grass and soybean groats. The results of observations during the research, soybean groats it was a favorite goat so that it can be spent in a short period of time that is around 5-10 minutes every time of feeding. The factors affect the feed consumption is the very nature of feed such as particles size, texture and scent. According to [12] that scent was an important factor of feed material. The animal can refused material feed given without feel because it was not fond of the scent. The other factors that were also affect of feed consumption like body sizes of animal, increasing feed consumption in line with increasing the size of animal [13].

Groats soybeans provided can be spent starting the first day until the eighth day collection of fodder though mixed with CPH fermentation, animal first was eat more groats soybeans and to be continued eating of CPH fermentation. This case is caused by the goats have selective properties in consuming the feed through the senses of smell so it takes longer for the adaptation of the new type feed is given. The selective nature of the goat in consuming the feed associated with the tendency of choosing a feed that is no fibrous. According to [14] that was the goats have the ability to choose the type of feed to be consumed and preferred types of feed without fibrous.

3.2. Characteristic of Semen Macroscopic

The test semen quality under macroscopic examination such as semen of volume, color, smell, consistency, and the degree of the acidity or pH. The result of the observation of macroscopic in of the characteristics of fresh EGB semen in this research is still categories good and in the normal range of especially on P₀ and P₁, while P₂ and P₃ relatively less than the normal range of such as are presented in Table 3.

Table 3. Average of macroscopic characteristic of EGB semen

Semen characteristic	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Volume (ml)	0.90	0.87	0.43	0.37
Color	cream-yellowish	cream-yellowish	cream-yellowish	yellowish
Odor	specific	specific	specific	specific
Consistency	moderately viscous-viscous	moderately viscous-viscous	dilute-moderately viscous	dilute
pH	6.95	7.03	7.50	7.79

Analyzed in the Reproduction laboratory, Animal Science Faculty, UGM

The semen volume of EGB on P0 and P1 were 0.90 ml and 0.87 ml, respectively, these volume are still in the normal range, relatively the same with the volume of EGB semen was reported by [15] with average 0.95 ml. The volume of goat semen has varies every collecting with range of 0.5-1.5 ml [16]. Semen color is still in the normal range which it is creamy to yellowish on all feed treatment, but if the semen color is white to pale and it would affect on other semen characteristics. The semen consistency for P0 and P1 had moderately viscous to viscous, while P3 the semen quality was relatively low marked with dilute consistency. Color, consistency and concentration have links to each others. The semen color more faded, and the spermatozoa concentration become decrease and to be followed by more dilute of semen [17].

Semen pH of P0 and P1 were 6.95 and 7.03, respectively, semen pH in this research is still in the normal range to the EGB. According to [18], good quality of semen had a ph with a little acid from 7.0 and in average of 6.7. Semen pH in this research is closed to previous studies of semen pH for EGB. The results of previous studies reported that pH ranges about 6.85-7.13 for semen EGB [19, 15]. P2 and P3 had pH above the range of goat pH in both of 7.50 and 7.79 which is leads to the alkalis. The reaction of alkalis in spermatozoa was often associated with the low quality and low fertility [20].

The low semen quality in macroscopic in P2 and P3 had relation with feed consumption because during feed consumption was not optimal especially feed concentrates which was the more portion of CPH fermentation. P1 had low portion of CPH fermentation and higher portion of soybean groats in concentrate, while P0 had not give CPH fermentation and given higher portion of soybean groats. Groats soybean is a source of high protein that affects on the formation of the semen quality. The analysis of chemical composition was groats soybean having high crude protein content reached 33.07%, to be supported by organic matter 89.93% and crude fat 7.72%. The chemical composition of fresh semen plasma had high protein content about 214 mg/100 ml [21] or 4.59% as component in semen plasm.

3.3.Characteristic of Semen Microscopic

Microscopic examination of goat semen is mass motions, individual motions, concentration, motility, viabilitas and abnormality spermatozoa. Table 4 is show average of characteristic for EGB semen microscopic.

Table 4. Average of microscopic characteristic of EGB semen

Semen characteristic	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Mass movement	+++	+++	++	++
Motility (%)	78.33	76.67	51.67	46.67
Concentration (ml)	2.40 x 10 ⁹	2.24 x 10 ⁹	1.01 x 10 ⁹	0.96 x 10 ⁹
Viability (%)	79.57	79.47	59.97	51.13
Abnormality (%)	5.59	5.93	10.63	16.77

Analyzed in the Reproduction laboratory, Animal Science Faculty, UGM, 2011

The mass movement of EGB semen of P₀ about +++ and P₁ ++, these treatment were includes the categories excellent marked with visible large waves, thick, dark, and many, like wisps of active black clouds move fast moving place. It is supported with the high motility of the sperm in P₀ and P₁ was 78.33% and 76.67%, this condition indicates that the individual movement of sperm is quite progressive. It is powered by a low sperm abnormality in P₀, P₁ and P₂ about 5% 5.93% and 10.63%, respectively, but P₃ had more over from standard recommendation reached 16.77%. The semen concentration in all the feed treatments was still in the normal range of 0.96-2.40 x 10⁶/ml, but it is still lower than the research results [15], 2.94 x 10⁶/ml. [22] recommend that the terms of semen deluted that it may had mass movement, individual movement more over 65%, abnormality spermatozoa in range 14-15%. While [8] reported that the minimum standards for the semen quality which can be used for artificial insemination (AI) was the minimum to contain 500 million cells/ml/mass movement/ejakulate, as well as 50% in both of semen viability and motility. The characteristics of EGB semen on this research especially P₀ P₁ and P₂ can be used to the AI program.

4. Conclusion

Based on the results of this research it can be concluded up as follows:

1. The addition of CPH fermentation in concentrates up to 20% for of EGB has not affect to the feed consumption.
2. The addition of CPH fermentation in concentrates up to 20% for of EGB has affect to decreasing of the semen quality in both of microscopic and microscopic.

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