



FEEDING OF GLUCOMANNANS AND ANTHOCYANINS COMBINATION IN THE CONTAINING MICROPARTICLE PROTEIN ON FAT DIGESTIBILITY AND FAT DEPOSITION ON BROILER CHICKEN

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ABSTRACT

This study aims to evaluate the effects of combination of glucomannan and anthocyanin in feeds containing protein microparticles on fat digestibility and fat deposition in broiler chickens. The 216 healthy 7-day-old broiler chicks strain CP 707 was used with treatment of glucomannan from porang tuber extract and anthocyanins from purple sweet potato extract. The study was arranged by a completely randomized design (CRD) with a factorial pattern with treatment consisting of 2 factors (A and B). In A, there were 3 glucomannan treatments, A1 (0%), A2 (0.05%) and A3 (0.1%), while in B contained 3 anthocyanin treatments, B1 (0), B2 (0.07%) and B3 (0.14%) with 3 replications. The measure was fat digestibility, percentage of abdominal fat and meat fat mass. The data were analyzed for variance at the 5% level and continued with Duncan's double test at the 5% level. The results showed that the addition of a combination of glucomannan and anthocyanin in the feed containing protein microparticles had an interaction ($P < 0.05$) on fat digestibility, meat fat mass and relative weight of abdominal fat in broiler chickens. The addition of a combination of 0.1% glucomannan and 0.14% anthocyanin (A3B3) in the feed containing microparticle protein was able to reduce fat digestibility, relative abdominal fat weight and meat fat mass in broiler chickens.

INTRODUCTION

The growing population in Indonesia leads to an increase of the communities nutritional needs, includes animal protein, especially in broiler chickens. Feed donates the largest cost in livestock production (Sari *et al.*, 2020). The use of feed additives aims to increase livestock productivity. However, feed additives in the form of

antibiotic growth promoter (AGP) has been banned by the government. Porang tuber extract (PTE) and purple sweet potato (PSPE) can be an alternative because it easily obtained in the field. Porang tubers contain 64.22% glucomannan (Setiawati *et al.*, 2017). The addition of glucomannan as a source of prebiotics stimulates the growth of lactic acid bacteria (LAB) which produce lactic acid and short chain fatty acid (SCFA), then the reduction of intestinal pH is reduce the population of pathogenic bacteria (Sa'diyah *et al.*, 2020). Glucomannan as dietary fiber reduce the cholesterol levels, improve digestive function and immune system (Rosalina & Cahyani, 2015).

Purple sweet potato extract contains anthocyanins as antioxidants, antimutagenic, and anticarcinogenic. Anthocyanins is an antioxidant which counteract free radicals and clumping of blood cells (Artadana *et al.*, 2016). The higher anthocyanin content in purple sweet potato can be used to bind body fat which then be excreted through the excreta and the absorbed fat in the body can be reduced (Nurazizah *et al.*, 2020). The anthocyanin content in purple sweet potato is 596 mg/L (Pratiwi & Priyani, 2019). The addition of purple sweet potato as a source of antioxidants has a positive effect on digestibility so that the nutritional needs of livestock can be met.

The feeds with protein sources such as fish meal and soybean meal are processed into microparticle sizes. Microparticles are feed ingredients whose particles have a size between 0.2 – 0.5 μm (Sari *et al.* 2019) so the nutrients is well absorbed and digested in the digestive tract because microparticle processing simplifies complex molecules into simple ones to facilitate nutrient absorption (Afriyanti *et al.* 2019). Feeds with microparticle size can affect feed digestion because the wider the particle size, the longer digestion in the intestines, thus slowing the rate of digestion (Suparti, 2018).

Addition of glucomannan from porang tuber extract in microparticle feed was used as a prebiotic. Glucomannan as a substrate can be utilized by beneficial bacteria in the digestive tract, so that the growth of beneficial bacteria in the small intestine can increase so as to produce SCFA which makes the digestive tract acidic. In addition, the addition of anthocyanins as phytochemicals to maintain the health of the digestive tract so that nutrient absorption can be optimal.

The acidic nature also supports the growth of beneficial bacteria and promotes digestive tract health. The acidic conditions in the digestive tract inhibit the work of the lipase enzyme to digest fat so that fat absorption in the small intestine is reduced. Low

pH changes inhibit lipase work to digest fat, resulting in reduced fat absorption in the intestine and low fat synthesis in the blood (Octavia *et al.* 2018). On the other hand, changes in pH increase protein absorption because proteases work at low pH conditions. Fat and protein are absorbed in the form of lipoproteins, so that protein is absorbed higher than fat. The amount of fat that is absorbed a little can provide quality meat to be healthy. This study aims to examine the effect of adding a combination of glucomannan and anthocyanin in feeds containing microparticle protein sources on fat digestibility, meat fat mass and percentage of abdominal fat in broiler chickens.

MATERIALS AND METHODS

Subject and material

Animals used as many as 216 DOC broiler chickens strain CP 707 with an average initial body weight of 44.05 ± 0.26 g. The feed given is composed of several feed ingredients (**Table 1**). The treatment materials used were porang tuber extract and purple sweet potato. Microparticle protein feed ingredients are soybean meal and fish meal. Equipment that supports the research is a set of fat test (sokhlet), electric stove, stirrer, 70% ethanol, fine filter paper, evaporator, and incubator.

Sample preparation

The PTE preparation is based on the procedure for making porang tuber extract by Perdinan *et al.* (2019) with a sample and solution ratio of 1:10. A 100 g of porang tuber flour was dissolved in 500 ml of distilled water and 500 ml of 70% ethanol (50:50), then stirred. The solution was boiled at 80°C for 30 minutes in waterbath with stirrer and then filtered with filter paper. The supernatan was stored at 10-15°C for 24 h. The pellet is dried in incubator. The dried pellet is grinded.

For the PSPE, a 100 g of purple sweet potato was added into 1 liter of 96% ethanol (1: 10). It was placed in an airtight dark room for 48 h and stirred once a day. It was filtered and placed in a beaker that was tightly wrapped with aluminum foil. Evapofeed using a tosator evapofeed device, then stored in refrigerator. The purple sweet potato extract was added to the feed according to the treatment.

The manufacture of fish meal and microparticle soybean meal refers to the method of Suthama and Wibawa (2018). The ingredients was smoothed, then 400 ml distilled water and 2% virgin coconut oil were added per 100 g of material. It was then placed for sonification for 60 minutes, filtered, and dried under the sunlight.

Table 1. Compositions and feed nutrient content

Feed Stuff	Composition (%)
Yellow corn	55.25
Rice bran	14.52
Soybean meal of microparticle	20.38
Fish meal of microparticle	9.00
CaCO ₃	0.30
Premix	0.21
Lysine	0.10
Methionine	0.20
Nutrient Content (%)	
Metabolizable energy (ccal/kg)**	3,000.05
Protein*	18.30
Crude fat*	4.51
Crude fiber*	5.89
Calcium*	1.09
Phosphor*	0.86

Sources: *Feed analysis result in Laboratory of Animal Nutrition Science, Faculty of Animal and Agriculture Sciences, Universitas Diponegoro (2021).

**Result are calculated based on Bolton's formula (1967).

Rearing chicken

Maintenance of broiler chickens starting from DOC until 7 days is feeded with commercial starter feed B-11S. Provision of feeds and drinking water on an ad libitum. At the age of 8 days, treatment feeds were given with a ratio of a mixture of commercial feeds and treatment feeds with a ratio of 25:75, at 9 days of age the ratio of 50: 50, at 10 days of age the ratio of 75:25 and at 11 days of age, 100% of the treatment feeds were given. Chickens were kept in litter cages for 35 days. The treatment was added to the feed once every morning.

Research design and treatment

A completely randomized design (CRD) with a factorial pattern consisting of 9 treatments with 3 replications and each consisting of 8 bird was used. Factor A with 3 glucomannan treatments sourced from PTE, A1 (0%), A2 (0.05%) and A3 (0.1%).

Factor B with 3 treatments of anthocyanins, B1 (0%), B2 (0.07%) dan B3 (0.14%). The treatment combinations were:

A1B1 = feed without PTE and PSPE

A1B2 = feed + PTE 0% and PSPE 0.07%

A1B3 = feed + PTE 0% and PSPE 0.14%

A2B1 = feed + PTE 0.05% and PSPE 0%

A2B2 = feed + PTE 0.05% and PSPE 0.07%

A2B3 = feed + PTE 0.05% and PSPE 0.14%

A3B1 = feed + PTE 0.1% and PSPE 0%

A3B2 = feed + PTE 0.1% and PSPE 0.07%

A3B3 = feed + PTE 0.1% and PSPE 0.14%

Determination of factor A levels based on the results of Perdinan *et al.* (2019) and factor B levels based on Avioleza (2019).

Data measurement

Parameters measured included crude fat digestibility, relative weight of abdominal fat and meat fat mass. Fat digestibility was measured by the total collection method of Fe₂O₃ indicator combination for 4 days at the age of 32-35 days. The 27 battery cages filled with one chicken from each replication were used fat digestibility measurement. The feed was mixed with 0.5% Fe₂O₃, then the red excreta were accommodated with a special mat placed at the bottom of the battery cage. The excreta were collected everyday by spraying with 0.2 N HCL in every 2 hours. The collected excreta were dried and crushed for fat content analysis with Soxhlet method. Fat digestibility is calculated with the formula from Krismiyo *et al.* (2020) as follows:

$$\text{Fat Digestibility (\%)} = (\text{fat intake} - \text{amount of fat excreta}) / \text{fat intake} \times 100\%$$

The fat content of the meat was determined from samples of meat from the chest, back, and thighs of all parts (top and bottom). The meat is separated from the bones, mixed and mashed then samples are taken for analysis of fat content. Fat content was analyzed by Soxhlet method. The fat mass of the meat was calculated using the formula according to Krismiyo *et al.* (2020) as follows:

$$\text{Meat fat mass (g)} = \text{meat fat content (\%)} \times \text{meat weight (g)}$$

Abdominal fat was taken from the abdomen and viscera, and weighed with a digital scale with an accuracy of 0.01 g. The percentage of abdominal fat was calculated using the formula according to Krismiyo *et al.* (2020) :

$$\text{Percentage of abdominal fat (\%)} = \text{fat weight (g)} / \text{body weight (g)} \times 100\%$$

Data analysis

The data obtained were then analyzed using analysis of variance at a significance level of 5%. If it has a significant effect, it will be continued with Duncan's double test with a significance level of 5%.

RESULTS AND DISCUSSION

The addition of a combination of glucomannan and anthocyanin in feeds containing protein microparticles showed an interaction ($p < 0.05$) on fat digestibility, fat content in meat and abdominal fat content in broiler chickens (**Table 2**).

Table 2. Fat digestibility, meat fat mass and percentage of abdominal fat in broiler chicken

Level of PTE	Level of PSPE			Average
	B1	B2	B3	
------(%)-----				
Fat digestibility				
A1	84.25 ^a	78.13 ^b	77.45 ^b	79.94
A2	76.55 ^b	77.57 ^b	75.05 ^{bc}	76.39
A3	76.26 ^c	72.16 ^{cd}	71.06 ^d	73.16
Average	79.02	75.95	74.52	76.49
------(g)-----				
Meat fat mass				
A1	30.56 ^a	30.43 ^a	27.59 ^a	29.53
A2	24.12 ^b	22.24 ^b	23.31 ^b	23.22
A3	23.93 ^b	21.60 ^b	17.47 ^c	21.00
Average	26.20	24.76	22.12	24.58
------(%)-----				
Percentage of abdominal fat				
A1	2.11 ^a	2.00 ^a	1.74 ^b	1.95
A2	1.65 ^b	1.64 ^b	1.36 ^c	1.55
A3	1.47 ^c	1.19 ^d	1.17 ^d	1.27
Average	1.74	1.61	1.42	1.59

Source: Different superscripts in the same row and column showed significant differences ($P < 0.05$)

Fat digestibility

Duncan's test results showed that the A3B3 treatment with a combination of 0.1% glucomannan and 0.14% anthocyanin resulted in the lowest crude fat digestibility. The low crude fat digestibility value in the A3B3 treatment with the addition of a combination of 0.1% glucomannan and 0.14% anthocyanin was due to the role of bile salt hydrolase (BSH) enzyme secretion which suppresses fat digestibility so that the fat digestibility value decreases. This is because the combined use of glucomannan as a prebiotic and anthocyanin as an antioxidant is effective in increasing beneficial bacteria in the digestive tract. Sa'diyah *et al.* (2020) which states that the addition of a combination of glucomannan and anthocyanins can stimulate the growth of LAB which produces metabolite products such as lactic acid and short chain fatty acids (SCFA) which can lower the pH in the intestine thereby reducing the population of pathogenic bacteria and increasing lactic acid bacteria. According to Krismaputri *et al.* (2019) that the mechanism for decreasing pH is caused by SCFA organic acids when in cells are able to dissociate or separate and then produce positive hydrogen ions (H⁺) as a result, cell pH decreases so that the digestive tract becomes acidic. Acidic environmental conditions cause an increase in lactic acid bacteria so that the amount of BSH enzymes produced also increases. The BSH enzyme produced by bile secretion will go down into the small intestine then with the fermentation of lactic acid bacteria will experience conjugation so that it comes out and is distributed in the digestive tract, the digestive tract becomes acidic and causes fat digestion to be hampered. According to the opinion of Harumdewi *et al.* (2018) that lactic acid bacteria play a role in the secretion of bile salt hydrolase (BSH) enzymes which make bile salts conjugated and not easy to emulsify fats so that the fat absorption process in the intestine is reduced and fat digestibility is low.

Meat fat mass

Duncan's test results showed that the A3B3 treatment with a combination of 0.1% glucomannan and 0.14% anthocyanin resulted in the lowest meat fat mass. This is because low fat mass of meat is associated with low fat digestibility. The use of extracts of porang tubers and purple sweet potatoes cause the lower or acidic intestinal pH due to the increase of lactic acid bacteria and inhibition of pathogenic bacteria .

According to Kirana *et al.* (2017) that acidic environmental conditions cause the growth of pathogenic bacteria to be inhibited and the performance of the lipase enzyme is limited so that fat digestibility is reduced and the fat content of meat decreases. The low fat mass results are also caused by the use of microparticle protein so that the nutrients in the feed are more easily absorbed besides the bacteria in the digestive tract making it easier to ferment so that it produces more SCFA and increases the population of lactic acid bacteria thereby lowering the pH in the intestine. According to Abdurrahman *et al.* (2016) that by giving probiotics and antioxidants it can increase lactic acid bacteria so that these bacteria are able to produce BSH enzymes which make bile salts conjugated and do not emulsify fats so that fat digestibility decreases and meat fat mass is also low. In addition, the fat mass of meat is influenced by the nutritional content in the feed. According to Berliana *et al.* (2020) that Fat content in poultry meat the higher, the cholesterol level of meat also high and vice versa.

Percentage of abdominal fat

Duncan test results show that the A3B3 treatment in Table 2. produces the lowest relative weight of abdominal fat. This is due to the presence of glucomannan in porang tuber extract as a prebiotic and anthocyanins in purple sweet potato extract as antioxidants. Probiotics and antioxidants in the feed are useful for improving animal health and maintaining the balance of microflora in the digestive tract and inhibiting the growth of pathogenic bacteria in the digestive tract of broiler chickens. Faradila *et al.* (2016) stated that the results of the metabolism of porang tuber extract and purple sweet potato extract can lower the pH in the intestine thereby suppressing the growth of pathogenic bacteria and increasing the growth of lactic acid bacteria so that nutrient absorption can be maximized. However, under acidic conditions it can reduce the activity of the lipase enzyme in digesting fat because the enzyme is active at a neutral pH, resulting in reduced fat absorption in the intestine.

According to Octavia *et al.* (2018), lactic acid bacteria produce lactic acid which results in acidic conditions in the digestive tract thereby reducing the work of the lipase enzyme to digest fat, resulting in reduced fat absorption in the intestine and low fat synthesis in the blood. In addition, the crude fiber content in the feed can also affect the weight of abdominal fat. Crude fiber from the feed after consumption will bind bile

acids when they arrive in the digestive tract, causing the function of bile to help the absorption of fat to be inhibited. According to Wu *et al.* (2020), bile acids that have been bound by crude fiber will be excreted from the body in the form of feces, resulting in a decrease in abdominal fat deposition.

CONCLUSION

Feeding of a combination of glucomannan from 0.1% porang tuber extract and 0.14% anthocyanin from purple sweet potato was able to reduce fat digestibility, meat fat mass and relative weight of abdominal fat in broiler chickens.

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