

## **Carcass and Tibia Bone of Broiler Chickens Fed *Amorphophallus companulatus* tubers Fermented by *Bacillus subtilis***

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**ABSTRACT:** The objective of the present study was to investigate the effect of fermented *Amorphophallus companulatus* (AC) tubers by *Bacillus subtilis* on the final weight, carcass weight, and tibia bone of broiler chickens. The feeding trial lasted for 35 days. One hundred and twelve eight days old male broilers of the *New Lohmann* strain were used for the study. There were four treatment groups and four replications with seven birds per replicate. The four treatments were 1) a control diet without FAC, 2) a diet with 5% FAC, 3) a diet with 10% FAC, and 4) a diet with 15% FAC. The variable observed were final weight, carcass weight, abdominal fat weight, percentage of carcass weight, length, and weight of tibia bone. The result showed that feeding FAC tubers meal did not affect ( $P>0.05$ ) final weight, carcass weight, the percentage of carcass weight, length, and weight of tibia bone. Feeding FAC tuber meal up to 15% had lower ( $P<0.05$ ) abdominal fat than feeding control dietary. In conclusion, fermented AC by *Bacillus subtilis* can be used up to 15% in broiler diets without adversely affecting carcass weight and tibia bone.

**Keywords:** *Amorphophallus companulatus*; *Bacillus subtilis*; Fermentation; Carcass; Tibia bone; Broiler chicken.

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## INTRODUCTION

*Amorphophallus companulatus* (AC) tubers can be used as feed alternative energy sources. AC grows wild in the forest. The tubers are not consumed (Yuzammi, 2009). This plant is a wild plant that grows in the forest and has not been widely cultivated in Indonesia (Santosa *et al.*, 2013). This plant has been cultivated and has a production of 50-80 tons/ha in India (Ravi *et al.*, 2009). The AC tuber contains 7.33% crude protein and 3570.60 kcal/kg gross energy (Koni *et al.*, 2017); 34 mg / 100 g phosphorus, 50 mg / 100 g calcium, and vitamin A 434 IU / 100 g (Ravi *et al.*, 2009); 0.46% crude fat, 32.1 mg / 100 g calcium, and 1.68% crude fiber (Chattopadhyay *et al.*, 2010).

The presence of oxalate is one of the major limiting factors in the utilization of AC which impart an acrid taste or cause irritation when food is prepared from that. Ingestion of foods containing oxalates has also been reported to have a caustic effect, exert irritations to the intestinal tract and cause absorptive poisoning (Rahman *et al.*, 2013). Root crops (*Dioscorea alata*, *Typhonium trilobatum*, and *Amorphophallus*) contain oxalic acid 0.45 - 0.78%. (Anbazhagan *et al.*, 2007) Calcium oxalate of AC in the form of raphide (Nugroho, 1996). A. Mueller tuber flour contains 6.24% calcium oxalate (Rosidah *et al.*, 2013), *Amorphophallus companulatus* (AC) tuber contains 318,510 mg/kg oxalate (Koni *et al.*, 2017b) Paul *et al.*, (2013) Oxalate, consumed in sufficient quantity, can bind with calcium (Ca) in the intestines and the blood to form insoluble Ca oxalate, which may lead to low serum Ca (Rahman *et al.*, 2013)

High oxalate feed affects the formation of calcium oxalate crystals in the rats' kidneys (Rosidah *et al.*, 2013). Rahman *et al.* (2011) stated that ruminants have a higher resistance to oxalates than monogastric because microorganisms digest oxalates in the rumen. (Rahman *et al.*, 2013) argued that to avoid harming livestock, the oxalate content in non-ruminant animal feed

should be less than 0.5% while it could reach 2% for ruminants.

Fermentation can reduce anti-nutrients and improve the nutrients content of feed ingredients (Steinkraus, 1994; Feng *et al.*, 2007; Chiang *et al.*, 2010; Ojokoh *et al.*, 2013), and increase nutrient digestibility (Hilkias *et al.* 2017). Five days fermented by *Bacillus subtilis* caused an increase of 25.54% crude protein, 2.63% crude fat, and a decrease of 5.23% crude fiber and 70.81% oxalate of *Phaseolus lunatus* (Adegbehingbe *et al.*, 2014). Fermentation with *Bacillus subtilis* on cotton seeds can improve flavor, and palatability, increase 3.42% crude protein, 14.58% crude fat, and decrease by 3,26% crude fiber. (Kanyinji and Sichangwa, 2014). Koni *et al.* (2017b) stated that the oxalate in AC tubers could be reduced up to 53.2% with fermentation by *Bacillus subtilis* for seven days at room temperature. Therefore, this study aimed to determine the carcass and tibia bones in broiler chickens fed with AC fermented with *Bacillus subtilis*.

## MATERIALS AND METHODS

### Fermentation process

AC was obtained from East Amarasi Village, East Amarasi District, Kupang Regency, East Nusa Tenggara. Tubers were cleaned with tap water to remove the soil on tuber peels. The tubers were sliced to  $\pm 7$  cm length and  $\pm 3$  cm thickness, sundried for  $\pm$  two days, and milled. *Bacillus subtilis* in solid form was obtained from Microbiology Laboratory Pusat Antar Universitas (PAU) Universitas Gadjah Mada. Dried tubers were as milled. AC tubers were fermented by the solid fermentation method. AC meal was inoculated with 20% *Bacillus subtilis* based on dry matter. AC meal was mixed with *Bacillus subtilis* until homogenous and then placed in a plastic bucket with a capacity of 8 kg as a silo, compacted, and fermented at room temperature for seven days.

### Experimental birds, design

A total of 112 eight-day-old male New Lohmann, MB-202 Platinum broiler

chickens were randomly allocated to 4 groups of 7 birds each, comprising four replicates per treatment. The birds were fed on the experimental diets from eight-day-old to 35 days old. Feed and water were provided ad libitum and different levels of fermented AC (FAC) in the broiler's diet as a treatment. The dietary treatments were: 1) a control diet without FAC, 2) a diet with 5%

FAC, 3) a diet with 10% FAC, and 4) a diet with 15% FAC. The chicks were fed with the treatment diets from d 8 to 35. Diets were formulated with crude protein 21.02 to 21.39% and metabolized energy 3027.17 to 3044.77 kcal/kg. The ingredient and chemical compositions of the experimental diets used in this study are shown in Table 1.

**Table 1.** Ingredients and chemical composition of dietary treatments

Ingredient	Dietary treatment (FAC (%))			
	0	5	10	15
Corn	53.00	50.50	48.00	45.50
Rice bran	13.00	10.50	8.00	5.50
Meat bone meal	7.35	7.35	7.35	7.35
Soybean meal	25.00	25.00	25.00	25.00
Vitamin and trace element premix <sup>1)</sup>	0.50	0.50	0.50	0.50
dl-Methionine	0.30	0.30	0.30	0.30
Lysine hydrochloride	0.60	0.60	0.60	0.60
Salt	0.25	0.25	0.25	0.25
FAC	0.00	5.00	10.00	15.00
Total	100.00	100.00	100.00	100.00
Calculated values				
Dry matter (%)	86.77	86.43	86.08	85.73
Crude protein (%)	21.39	21.29	21.18	21.07
Metabolizable energy (kcal/kg)	3027.20	3044.80	3062.40	3079.90
Crude fiber (%)	4.25	4.06	3.86	3.66
Crude fat (%)	4.24	3.97	3.71	3.45
Calcium (%)	0.91	0.95	0.98	1.02
Phosphorus (%)	0.48	0.48	0.48	0.48
Lysine (%)	1.02	1.01	1.00	1.00
Methionine (%)	0.53	0.53	0.52	0.52
Oxalate (mg/kg)	0.00	4.84	9.69	14.53

Note: FAC: Fermented *Amorphophallus companulatus*

On the 35th day, two chickens were taken randomly per experimental unit to be slaughtered, and carcass weight and abdominal fat were measured. The left tibia was taken and then cleaned of soft tissue, put in boiling water for 10 minutes, cleaned of bone covering, and left for 24 hours at room temperature to dry. The length of the tibial bone is measured in cm, and its weight is weighed in grams.

**Statistical analysis**

A one-way ANOVA procedure statistically analyzed all the data. Duncan's

multiple range test was further performed when significant differences were found ( $P < 0.05$ ). Statements of significance were based on  $P < 0.05$  (Gasperz, 2006)

**RESULT AND DISCUSSION**

The feed intake, slaughter weight, carcass, and tibia weight of the broiler at 8-35 days old are presented in Table 2.

The results showed that FAC did not affect ( $P > 0.05$ ) feed intake, slaughter weight, and carcass weight of chickens. The same feed intake in all treatments was

possible because the metabolizable energy of feed was almost the same, and the fiber content was low. It could still be tolerated by broilers (Table 1). The difference in metabolizable energy content in poultry feed affects the amount of feed intake (Ariesta *et al.*, 2015). High crude fiber content has a negative effect on feed consumption

(Binowo *et al.*, 2019). Although FAC contains oxalate, it does not negatively affect chicken feed consumption. The presence of anti-nutrient factor components such as protease inhibitors, alkaloids, phytates, and oxalates will reduce feed consumption and suppress livestock growth (Ferket & Gernat, 2006).

**Table 2.** Feed intake, Slaughter weight, carcass, abdominal fat and tibia weight, tibia length of broiler that fed fac fermented by *Bacillus subtilis*

Parameter	FAC (%)				SEM	P-value
	0	5	10	15		
Feed intake (g)	2994.28	2962.78	2936.0	2930.10	11.925	0.213
Slaughter weight (g)	1500	1493.38	1480.38	1476.13	6.001	0.470
Carcass weight (g)	1085.25	1072.63	1068.13	1054.75	4.74	0.143
Abdominal fat weight (g)	10.73a	7.72b	6.82b	6.66b	0.55	0.012
Carcass percentage (%)	72.45	73.49	72.23	75.58	0.19	0.06
Tibia weight (g)	4.25	4.25	4.2	4.09	0.04	0.502
Tibia length (cm)	8.68	8.13	8.45	8.45	0.12	0.497

Note: FAC: fermented *Amorphophallus companulatus*, SEM: Standard error of the mean, p: probability, <sup>ab</sup>Means in the same row without a common letter are different at P<0.05

The weight gain of White Leghorn chickens aged six weeks was reduced by an increase in oxalic acid in the feed, namely 495, 464, 460, and 392 g for each application of 0, 0.25, 0.50, 1% of oxalic acid, respectively (Robertson, 2015). The feed intake of chickens fed FAC was higher than chickens fed AC without fermentation, which was 2083.61 + 42.49, 1946.15 + 53.12, and 1824.43 + 50.42 g/bird AC 5, 10, and 15%, respectively (Koni *et al.*, 2019). The same slaughter and carcass weight followed the same feed intake in all treatments. The result showed that FAC had no significant effect (P <0.05) on the percentage of broiler chicken carcasses. This is because birds fed FAC have low body weight, and the resulting carcass weight is low; on the other hand, those fed fermented feed have high body weight followed by high carcass weight, resulting in a percentage that does not look different between treatments. Soeparno (2009) states that carcass usually increases with increasing live weight but that the percentage of non-carcass parts such as

blood and vital organs decreases. There was an increase in the carcass percentage of broiler chickens fed *Bacillus* spp by 4.64% compared to chickens fed control feed (Hidayat *et al.*, 2016). Osei and Duodu (1988) stated that an increase in the use of naturally fermented cassava peels for three days had a better carcass percentage, namely 76 ± 15, 75 ± 2, 76 ± 16, 76 ± 18% when using 0, 0.5, 1 and 1.5% in broiler chicken feed. The percentage of the carcass in this study ranged from 72.15 to 73.45%. The percentage carcass of broiler Lohmann strain broiler was from 63.85 to 66.16% (Sibrani *et al.*, 2014)

The results showed that FAC had a significant effect (P <0.05) on the abdominal fat of broiler chickens. The abdominal fat in control birds was significantly higher (P <0.05) than in the group fed FAC. The *Amorphophallus* sp. causes this contains carbohydrates that are suitable for lysing fat. *Amorphophallus companulatus* has a low glycemic index so that carbohydrates consumed are slowly stored in the form of fat (Faridah, 2005); *Amorphophallus* has

starch which is suitable for people with obesity and hyperlipidemia (Suriya *et al.*, 2017).

The tibia length in this study ranged from 7.30 to 8.68 cm. Statistical analysis showed that FAC had no significant effect ( $P > 0.05$ ) on the length and weight of tibia broiler chickens. Birds fed a control diet have tibia lengths the same as those fed *Amorphophallus companulatus* fermented by *Bacillus subtilis*. This is because broiler chickens can tolerate the same consumption of calcium and oxalate in the fermentation treatment so that it does not significantly interfere with the calcium absorption process.

The fermentation by *Bacillus subtilis* produces oxalate decarboxylase so that the minerals in the feed are available for livestock; besides, the fermentation process can produce the vitamins needed in the calcium metabolism process for bone formation. Microorganisms used in the fermentation process can synthesize vitamins, such as D, C, and K, which affect calcium metabolism for bone formation. (Mutuş *et al.*, 2006).

Calcium consumption in control birds was 0.78 g/bird/day while those fed FAC of 0.74 to 0.77 g/bird/day and oxalate intake ranges from 0.41 to 1.21 mg/bird/day. Koni *et al.* (2021) reported that oxalate content in dried tubers of *Amorphophallus* was higher than in fermented tubers, which were 0.38mg/kg and 0.097mg/kg, respectively. *Amorphophallus* sp. tuber consists of oxalate, but other fermented products can help in the metabolic process of minerals used for bone formation.

Fermentation products increase calcium retention, thereby affecting the increase in tibia weight in broilers, namely 1.43 g in control to 1.53 g in fermented products (Han *et al.*, 2015). Applegate and Lilburn (2002) stated that the tibia length in male and female broilers at the age of 21, 28, and 35 days were in males 6.72, 8.10, 9.42 cm in females, 6.8, 8.04, 9.05 cm, while tibia weight in males 2.36, 4.13, 6.09 g and in females 2.29, 3.37 and 4.96 g.

## CONCLUSIONS

FAC can be given up to 15% in broiler chicken feed without harming slaughter weight, carcass weight, carcass percentage, tibia length, and weight. Using fermented *Amorphophallus companulatus* tuber up to 10% in broiler ration can reduce abdominal fat.

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