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*by* Harnentis2 Harnentis2

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## 1 The utilization of different binders for pelleted native chicken ration based on coconut meat waste supplemented with mannanolytic thermophilic bacteria and thermostable mannanase: a physical characteristic of pelleted native chicken ration

Harnentis<sup>1</sup>, Yetti Marlida<sup>1</sup>, and Robi Amizar<sup>1</sup>

<sup>1</sup> Department of Animal Nutrition and Feed Technology, Faculty of Animal Science, Andalas University, Padang, West Sumatera, Indonesia.

**Abstract.** This research aim was to know the influence of addition of binders (bentonite, tapioca flour, gambier liquid waste, and onggok) on physical characteristics of pelleted native chicken ration based on coconut meat waste supplemented with mannanolytic thermophilic bacteria and thermostable mannanase. A completely randomized design was applied in this experiment with five treatments and five replicates. Moisture content, angle of response, bulk and compacted bulk densities, specific gravity, compaction rate, and pellet durability were determined. Data were analyzed by analysis of variance according to completely randomized design and Duncan's Multiple Range Test. Data indicated that moisture content, bulk density, compact bulk density and pellet durability affected significantly by binders, while angle of response, specific gravity and compaction rate had no affected by the presence of binders. Moisture content, bulk density, compacted bulk density, pellet durability of ration with binders were significantly different with ration without binder. Ration with gambier liquid waste binder had the highest value for bulk density and compacted bulk density, i.e.  $1.36 \text{ g/cm}^3 \pm 0.011$  and  $1.46 \text{ g/cm}^3 \pm 0.001$  respectively, while the value of pellet durability in ration with bentonite and tapioca flour binders were higher than others ( $98.63 \pm 0.11$  and  $98.49 \pm 0.05$ ). It is concluded that the addition of binders in native chicken ration based on coconut meat waste supplemented with mannanolytic thermophilic bacteria and thermostable mannanase pelleted were able to improve the physical characteristic of native breed chicken ration.

**Keywords** – pelleted native chicken, coconut meat waste, mannanolytic thermophilic bacteria, thermostable mannanase.

### 1. Introduction

Indonesian local poultry include native chickens whose existence is well known to the public have a diverse appearance. Consumer demand for native chicken needs already very high. This can be seen from the increase in population growth and demand for native chickens every year [1]. The problem in developing domestic chicken is the high cost of feed. Feed is the biggest component and the success factor of a farm business. The quality and quantity of animal feed is often an obstacle that must be faced in an effort to meet basic life needs, growth, production and reproduction of livestock. For this reason, we need to find alternative sources of feed ingredients that have sufficient nutritional value, low prices, easy to obtain and safe for consumption by livestock.



Coconut meat waste is a by-product of coconut milk processing industry that has the potential to be used as animal feed, especially native chicken, because the production is abundant and has sufficient nutritional value. The use of coconut meat waste as animal feed, especially poultry, is still limited, because it is not palatable, voluminous and high in hemicellulose fiber content namely mannan and galactomannan. Many studies show that the administration of probiotics and enzymes can improve feed quality, so that it can reduce the limited use of feed, especially native chicken feed. A decrease in the hemicellulose content of coconut meat waste from 21.54% to 17.72% BK, using 800 U / kg of thermostable mannanase enzymes [2]. Supplementation of thermophilic mannanolytic bacteria (*Bacillus* sp. SM-1.4) 1010 CFU/kg in broiler rations based on coconut meat waste-based on can improve intestinal histomorphology, nitrogen retention, and metabolizable energy [3].

Currently poultry feed is switched from mash to pellet form, because it considered very effective. It is generally accepted that compared to mash, the feeding of pellets improves feed consumption [4], reduce the amount of wasted feed [5], make feed more homogeneous, reduce the time and use of energy to eat [6], besides giving it relatively easy and not dusty, increasing the bulk density of feed, improving feed flow ability, prolonging storage. and facilitate transportation [7].

Thermostable enzymes and thermophilic bacteria are needed in a feed product if the process of making these feeds uses high temperatures such as in making pellets. The physical form of pellets is strongly influenced by the type of material used, the size of the printer, the amount of water, pressure and the method after processing and the use of binders to produce pellets with a strong, compact and sturdy structure so that the pellets are not easily broken. The adhesive serves as an adhesive between all raw materials so that the feed that is made becomes more compact and stable. The adhesive commonly used by animal feed factories is synthetic adhesives such as bentonite, and lignosulfonate [8]. Synthetic adhesives are relatively expensive so we need to look for alternative adhesive materials from cheap local feed ingredients such as tapioca flour, onggok, and gambir liquid waste.

Besides being cheap, easy to obtain, it is also a natural material that tends to be safer to add to feed. The aim of this research is to produce pelleted native chicken ration based on coconut meat waste supplemented with *Bacillus* sp. SM-1.4 and thermostable mannanase using low cost starch based natural binders and synthetic binder (bentonite clay) leads to analyze and choose best pellets keeping in view the physical properties of pellets. Selection of binder can play the important role to achieve the same desired properties of pellets without steam conditioning and preheating of biomass material.

## 2. Materials and methods

**Bacteria Preparation.** a Pure culture of *Bacillus* sp. SM-1.4 isolated from hot springs South Solok [9], grown in media nutrient broth (NB) and then incubated at 60°C, for 24 hours. Once it is done counting the number of bacteria by a method using the Standard Plate Count Agar, to obtain the number of bacteria 1010 CFU/ml. The bacteria are then stored in a bottle.

**Production of Enzymes.** Mannanase thermostable enzyme produced by thermophilic mannanolytic bacteria (*Bacillus* sp. SM-1.4) [9]. *Bacillus* sp. SM-1.4 were grown in a medium containing (g/L). 40.9 copra meal, NH<sub>4</sub>NO<sub>3</sub> 0.5, Na<sub>2</sub>HPO<sub>4</sub> 7.54, NaH<sub>2</sub>PO<sub>4</sub> 2.32, MgSO<sub>4</sub>.7H<sub>2</sub>O 0.4, FeSO<sub>4</sub>.7H<sub>2</sub>O 0.02, CaCl<sub>2</sub>.2H<sub>2</sub>O 0.06, yeast extract (pH 7.0). Inoculum used for the production of the enzyme is *Bacillus* sp. SM-1.4 were cultivated in NB medium at a temperature of 60°C on a shaker water bath (120 rpm) for 12 hours. Enzymes produced using Minifors tool Benchtop Bioreactor with a capacity of 5 L. Medium inoculated with 10% inoculum (v/v) and incubated at 60°C for 18 hours, then centrifuged at a speed of 5,000 rpm for 5 minutes. The supernatant obtained is used as a crude enzyme in this study after the first test of enzyme activity.

**Experimental design.** This research was carried out using experimental method using Complete Design with 5 treatments and 5 replications. The treatment consists of: D0: diet without binder; D1: diet with bentonite binder 1.5%; D2: diet with tapioca flour binder 1.5%; D3: diet with gambier 1.5% liquid waste binder. D4: diet with 1.5% onggok binder. Diets were formulated in iso-nitrogenous. The nutritional content of the ration adjusted to the needs of protein for the growth of native chicken,

which is 19% and the metabolic energy is 2850 kcal/kg [10] as presented in Table 1. The five diets imposed are described in Table 1.

**Table 1.** Composition of experimental diets.

Ingredients (%)	Experimental Diets				
	D0	D1	D2	D3	D4
Yellow corn	23.0	23.0	23.0	23.0	23.0
Coconut meat	30.0	30.0	30.0	30.0	30.0
waste	9.8	9.8	9.8	9.8	9.8
Fish meal	19.7	19.7	19.7	19.7	19.7
Soybean meal	11.5	11.5	11.5	11.5	11.5
Japfa BRI	5.5	5.5	5.5	5.5	5.5
Rice bran	0.5	0.5	0.5	0.5	0.5
Top mix					
Total	100	100	100	100	100
Binder	Without binder	Bentonite (1.5%)	Tapioca flour (1.5%)	Gambir liquid waste (1.5%)	Onggok (1.5%)
<b>Calculated</b>					
Crude protein (%)	19.01	19.01	19.01	19.01	19.01
ME (kcal/kg)	2857.00	2857.00	2857.00	2857.00	2857.00
Crude fiber (%)	9.39	9.39	9.39	9.39	9.39
Lipid (%)	9.49	9.49	9.49	9.49	9.49
Ca (%)	0.89	0.89	0.89	0.89	0.89
P <sub>available</sub> (%)	0.42	0.42	0.42	0.42	0.42
Lysine (%)	1.29	1.29	1.29	1.29	1.29
Methionine (%)	0.49	0.49	0.49	0.49	0.49
Tryptophan (%)	0.24	0.24	0.24	0.24	0.24

### 2.1. Binder preparation

Binder used in this research is bentonite was purchased from market Bogor. Tapioca flour bought in the traditional market Padang, West Sumatera. Gambir liquid waste is obtained from traditional gambir production in Payakumbuh, West Sumatera. Onggok is obtained by separating starch from cassava. Cassava after being grated then added water is homogenized then squeezed using gauze until the starch runs out. The remaining starch (onggok) is dried in the sun for 2 days, then milled with a blender. For this purpose, each binder of 150 grams was boiled at 80°C in 5.0 liters of water. Binders are ready to be mixed with feed ingredients.

### 2.2. Preparation of pellets production

Coconut meat waste is obtained in the traditional market, dried for 12 hours by help of the sunlight, then finely ground. Other ration composition materials such as corn, soybean meal, and fish are also finely ground using a hammer mill to flour. Coconut meat waste before being mixed constituents of diet others added a mixture of crude enzyme mannanase with a dose of 800 U/kg of coconut meat waste and thermophilic bacteria (*Bacillus* sp. SM-1.4) 1010 CFU/kg diet [11]. The preparation of diets using materials and compositions as shown in Table 1, and formed pellet using 3 mm die with a length of 0.7-1.3 cm and then dried for 8 hours, then tested the physical quality of pellets in the Laboratory of Technology and Feed Industry, Faculty of Animal Science.

### 2.3. Stack angle [12]

Stack angle measurement is done by dropping 500 grams of material at a height of 32.5 cm through a funnel on a flat plane. As a flat plane, white paperboard is used. Stack diameter is twice as high as

the fall of material, while to measure height is done with calipers. The weight of the material used is 500 grams.

#### 2.4. Bulk density [13]

Bulk density is calculated by inserting a material with a certain weight into a 500 ml measuring cup with the help of a funnel. In addition, avoidance of shocks in the measuring cup during measurement. Bulk density expressed in units of  $\text{g} / \text{cm}^3$ . Bulk density was calculated as the ratio of the weight of the material (g) to the volume of space ( $\text{cm}^3$ ).

#### 2.5. Compacted bulk density [13]

Compacted bulk density determined in the same way as bulk density determination, but the volume of material is read after the compaction process by shaking the measuring cup by hand until the volume does not change again. Stack compaction density is expressed in units of  $\text{g} / \text{cm}^3$ . Compacted bulk density was calculated as the ratio of the weight of the material (g) to the volume of space after compacting.

#### 2.6. Specific gravity [13]

The Specific gravity is measured using the Archimedes Law principle, that is by measuring the change in volume in the measuring cup (500 ml) after added the determined amount of distilled water (200 ml) and material known to its mass (100 grams) into the measuring cup. Stirring is done in a measuring cup to accelerate the loss of air between the particles of the ration during the measurement. Changes in the volume of distilled water are the actual volume of material. Specific gravity was calculated as the ratio of the weight of the material (g) to change in the volume of distilled water ( $\text{cm}^3$ ).

#### 2.7. Pellet durability

Pellet durability was determined in a Holmen Pellet Tester. Clean pellet samples (200 g), with no fines, were rapidly circulated in an air stream around a perforated test chamber for 30 s. Fines were removed continuously through the perforations (2 mm in diameter) during the test cycle at 60 milbar. After the test cycle, the subject pellets were ejected and weighed manually. The pellet durability index (PDI) was calculated as the ratio of retained pellet weight after agitation to initial pellet sample weight.

#### 2.8. Pellet hardness

Pellet hardness was tested in a Stable Micro Systems Texture Analyzer using the method described [14]. Individual pellets a pressure piston and a bar, and by increasing pressure applied by means of the pressure piston, the force (Newton) needed to break the pellets was determined.

#### 2.9. Statistical analysis

The data obtained were analyzed statistically using diversity analysis according to Completely Randomized Design [15]. The differences between treatments were tested using Duncan's Multiple Range Test (DMRT).

### 3. Results and discussion

#### 3.1. The effect of treatment on the moisture content of pelleted native chicken ration

The effect of binders on the moisture content of pelleted native chicken ration presented in Table 2. The moisture content of pelleted native chicken ration significantly ( $P < 0.05$ ) affected by binders. The moisture content of pellet with gambir liquid waste significantly higher than other binders. The moisture content of pellet with bentonite had no significant with the without binder, but significantly ( $P < 0.05$ ) lower than other binders. The highest moisture content is obtained on pellets with gambier liquid waste binder, while the lowest water content is seen on pellets with bentonite and without

binder. The high level of moisture content of pellet with gambier liquid waste binder is caused due to gambier liquid waste containing tannin which has a stronger binding strength than other binders so that it inhibits the release / release of water that is between the particles of the material when it is dried in the same time.

Low water content in pellet with bentonite binder compared to other binders caused by heating on bentonite before being mixed with other feed ingredients, causes bentonite to be activated so that it can remove water molecules contained in the pellets. Bentonite before used must be activated first because in the initial state it only has the lowest adsorption ability. Activation of bentonite with the addition of hot water aims so that water is bound between molecule in the pellet ration can evaporate, so the porosity increases. Bentonite which binds to hot water can increase the ability of its absorption so can reduce the moisture content in the pellets. While low moisture content in pellet without binder caused the pores between the particles of the material are not tightly bound so that the water will be more volatile when drying.

**Table 2.** The mean of physical characteristic of pelleted native chicken ration in each treatment.

Variables	Treatments				
	D0	D1	D2	D3	D4
Moisture content (%)	8.34 ± 0.06 <sup>c</sup>	8.48 ± 1.02 <sup>c</sup>	9.40 ± 0.91 <sup>b</sup>	10.75 ± 3.60 <sup>a</sup>	9.05 ± 1.25 <sup>b</sup>
Angle of response (°)	32.05 ± 3.28	33.06 ± 1.69	32.82 ± 1.92	34.40 ± 2.12	32.88 ± 1.95
Bulk Density (g/cm <sup>3</sup> )	1.28 ± 0.004 <sup>d</sup>	1.31 ± 0.004 <sup>c</sup>	1.34 ± 0.005 <sup>b</sup>	1.36 ± 0.011 <sup>a</sup>	1.31 ± 0.004 <sup>c</sup>
Compacted Bulk Density (g/cm <sup>3</sup> )	1.38 ± 0.04 <sup>c</sup>	1.42 ± 0.04 <sup>b</sup>	1.45 ± 0.03 <sup>a</sup>	1.46 ± 0.01 <sup>a</sup>	1.41 ± 0.04 <sup>b</sup>
Specific gravity (g/cm <sup>3</sup> )	1.26 ± 0.03	1.32 ± 0.07	1.35 ± 0.08	1.38 ± 0.09	1.40 ± 0.11
Hardness (N/cm <sup>2</sup> )	173.84±1.59 <sup>b</sup>	237.43±2.7 <sup>a</sup>	213.43±2.13 <sup>a</sup>	217.33±1.41 <sup>a</sup>	226.81±1.30 <sup>a</sup>
Durability (%)	95.82 ± 0.23 <sup>c</sup>	96.61±0.11 <sup>b</sup>	98.49±0.05 <sup>a</sup>	98.63±0.11 <sup>a</sup>	96.86±0.26 <sup>b</sup>

: <sup>abc</sup> The mean with different superscripts at the same row differs statistically (P <0.05).

### 3.2. Effect of treatment on stack angle of pelleted native chicken ration

The effect of binders on the of stack angle of pelleted native chicken ration presented in Table 2. The use of different binders in pelleted native chicken ration had no significant (P >0.05) to the stack angle of pelleted native chicken ration. The value of the bulk angle in the research ration ranged 32.05°-34.40°. Bulk angle value on pelleted native chicken based on coconut meat waste supplemented with thermophilic bacteria and thermostable mannanase in this study included in the category of easy rations that are easy to flow on a sloping plane or a certain height that is, at the bulk angle range of 30-38° [16]. The using binders (bentonite, tapioca, gambier liquid waste and onggok) and without binder does not show an effect on the bulk angle of pelleted native chicken ration. The size of the bulk angle is affected by particle size, shape and characteristics, moisture content, specific gravity and bulk density [12]. Particle size affects bulk angle, ie the smaller the particle size, the higher the bulk angle [17]. Pellets that have a high bulk angle will reduce flow rate of pellet and will cohere so that the pellets are not free to move [18]. Pelleted ration can be categorized in solid form ration, where solid form rations have bulk angles ranging between 20° and 50° [12]. The value of the bulk angle in pelleted ration of native chicken in this study not much different that reported [19] with bulk angles values of commercial ration and pelleted basal ration of starter broiler with tapioca, bentonite and onggok binders which ranges from 33.43°-35.14°.

### 3.3. Effect of treatment on bulk density of pelleted native chicken ration

The effect of binders on the bulk density of pelleted native chicken ration presented in Table 2. The bulk density of pelleted native chicken ration significantly (P <0.05) affected by binders. Addition of different binder in native chicken ration significantly (P <0.05) affected bulk density pelleted native chicken ration based on coconut meat waste supplemented with thermophilic bacteria

and thermostable mannanase. Bulk density of pelleted native chicken ration with liquid gambier waste binder significantly ( $P < 0.05$ ) higher compared to others and without binder, then followed by pellet with tapioca flour binder. Addition of bentonite and onggok binders had no significant ( $P > 0.05$ ) to bulk density and the lowest obtained without a binder. The high density of pelleted ration with gambier liquid waste is caused by moisture content of pelleted ration is higher than that with other binders. The value of bulk density in this study getting lower with increasing moisture content, because the constituent ingredients of pellets will swell with higher moisture content causing the volume of space needed to be greater [20]. Materials that have low bulk density ( $< 0.45 \text{ g/cm}^3$ ) takes time to flow in a vertical direction for longer, on the contrary with materials that have a larger bulk density ( $> 0.5 \text{ g/cm}^3$ ) [13]. The results of bulk density in this study showed that the use of bentonite, tapioca flour, gambier liquid waste, onggok and without binder had a bulk density above  $0.5 \text{ g/cm}^3$ . The higher value of bulk density causes the volume of space loaded by pellets to be smaller. Materials with high bulk density can save costs for packaging and storing materials [21]. The bulk density of pelleted native chicken ration was higher than that reported [22], where the value of bulk density on synthetic binders pellets obtained was  $0.64\text{-}0.66 \text{ g/cm}^3$ .

#### 3.4. Effect of treatment on compacted bulk density of pelleted native chicken ration

The effect of binders on the compacted bulk density of pelleted native chicken ration presented in Table 2. Compacted bulk density of pelleted native chicken ration significantly ( $P < 0.05$ ) affected by binders. Compacted bulk density on pellets with gambier liquid waste binder was not significantly different ( $P > 0.05$ ) with tapioca flour binder, but significantly ( $P < 0.05$ ) higher compared to other binders. Pellets with bentonite adhesives give no significant effect ( $p > 0.05$ ) on stack compaction density and the lowest is obtained on pellets without binders. The difference between the value of compacted bulk density is caused by pellet water content [23], specific gravity, also influenced by the intensity and method of compaction which is conducted. Compaction on feeds with high specific gravity will increase compaction density [24]. The higher the value of compacted bulk density, the smaller the volume of space occupied by the pellet. The value of compacted bulk density on pelleted native chicken ration based on coconut meat waste supplemented with thermophilic bacteria and thermostable mannanase higher than that reported [22], which is equal to  $0.69 - 0.70 \text{ g/cm}^3$  in broiler ration pellets with synthetic binder.

#### 3.5. Effect of treatment on specific gravity of pelleted native chicken ration

The effect of binders on the of specific gravity of pelleted native chicken ration presented in Table 2. The use of different binders in pelleted native chicken ration had no significant ( $P > 0.05$ ) to the specific gravity of pelleted native chicken ration. The average specific weight of local chicken pellets with a binder of  $1.26 \pm 0.03 - 1.40 \pm 0.11 \text{ g/cm}^3$ . There is no influence of different binders on specific gravity of pelleted native chicken based on coconut meat waste supplemented with thermophilic bacteria and thermostable mannanase caused by compaction in the same pellet machine so that the space between particles is not different [25]. Rations consisting of particles of different specific gravity are large enough that this mixture is unstable and tends to be easily separated again [13]. Specific gravity values in the pellet ration in this study indicate level equal level of convenience in transportation and storage space capacity. The specific gravity of pellets in this study was not different from that reported [19], which ranged from  $1.27$  to  $1.40 \text{ g/cm}^3$  on starter broiler ration pellets with bentonite, tapioca and onggok binders, while [26] reported that the average specific density of pellets with lignosulfonate synthetic and bentonite binder were  $1.37 \text{ g/cm}^3$  and  $1.40 \text{ g/cm}^3$ .

#### 3.6. Effect of treatment on hardness of pelleted native chicken ration

The effect of binders on the hardness of pelleted native chicken ration presented in Table 2. The hardness of pelleted native chicken ration significantly ( $P < 0.05$ ) affected by binders. The average pellet hardness of pelleted native chicken ration with different binders ranged from  $173.84 \pm 1.59 - 237.43 \pm 2.7 \text{ N/cm}^2$ . Using the different binders was significantly ( $P < 0.05$ ) improved pellet hardness.



Pellet hardness in different binders, resistance to pellet breakdown due to pressure in bulk bins will also increase. Hardness of pellets with bentonite, tapioca flour, gambier liquid waste, and onggok binder significantly ( $P < 0.05$ ) higher than pellets without binders. This is probably influenced by the starch content of tapioca flour and onggok binder, while gambier liquid waste contained tannin which function as binder. That pellet hardness in pelleted broiler ration based on wheat and soybean meal range between 31.4 – 42.8 N [27].

### 3.7. Effect of treatment on of durability index of pelleted native chicken ration

The effect of binders on the durability index of pelleted native chicken ration presented in Table 2. Durability index of pelleted native chicken ration significantly ( $P < 0.05$ ) affected by binders. Durability index of pelleted native chicken ration using different binders ranged from 98.50% - 99.12%, the highest obtained in ration with gambier liquid waste and tapioca binders, then followed by pelleted ration with bentonite, onggok and without binder. Durability index on pellet with gambier liquid waste binder had no significantly different ( $P > 0.05$ ) with tapioca binders, but significantly ( $P < 0.05$ ) higher than the durability of pelleted ration with bentonite, onggok and without binders. This is due to gambier liquid waste containing tannin which has a higher adhesion than pellets with bentonite, onggok and without binder. while tapioca flour has starch with a higher amilopectin content than amylose. Pelleted native chicken ration based on coconutmeat waste supplemented with mannanolytic thermophilic bacteria and thermostable mannanase in this study had a high durability index, this may also caused by the work of the thermostable mannanase enzyme which breaks down mannan in coconut meat waste to produce mannosa which in turn increase gelatinization when mixing with hot binder (50-60°C) before pelleting Pellet durability is considered high when the calculated value is above 80%, while if the value is between 70% and 80%, and low when the value is below 70% [28]. The results of durability index pelleted native chicken ration in this study were higher than those reported [29], the shrimp feed pellet ration used 5% CMC binder, 5 and 10% tapioca binder, and 5% seaweed ranged between 92.26 - 94.44%.

## 4. Conclusion

It was concluded that using binders (bentonite, tapioca flour, gambier liquid waste, and onggok) could improved the physical quality of pelleted native chicken ration based on coconut meat waste supplemented with thermophilic bacteria and thermostable mannanase, while tapioca flour and gambier liquid waste showed best physical quality for pellet binders.

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