Chemical Content and In Vitro Digestibility of Broiler Litter Fermented at Different Ripen Time

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Chemical Content and *In Vitro* Digestibility of Broiler Litter Fermented at Different Ripen Time

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ABSTRAK

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Penelitian bertujuan untuk mengkaji pengaruh lama fermentasi litter ayam terhadap kandungan kimia dan nilai kecernaan secara *in vitro*. Penelitian menggunakan Rancangan Acak Lengkap dengan 4 perlakuan dan 4 ulangan, perlakuan tersebut adalah T0 = tanpa fermentasi; T1 = litter ayam fermentasi selama 3 minggu; T2 = litter ayam fermentasi selama 6 minggu dan T3 = litter ayam fermentasi selama 9 minggu. Parameter yang diamati yaitu kandungan kimia dan nilai kecernaan litter ayam fermentasi. Lama fermentasi yang berbeda mempengaruhi kandungan kimia litter ayam fermentasi yaitu kadar air, lemak, BETN dan TDN, namun tidak mempengaruhi kadar abu dan kadar serat. Lama fermentasi yang berbeda mempengaruhi nilai kecernaan bahan kering, kecernaan protein, kecernaan fraksi serat (ADF, NDF, Hemiselulosa), namun tidak mempengaruhi nilai kecernaan bahan organik, konstentrasi VFA, konsentrasi NH3 dan prouksi protein total litter ayam. Berdasarkan hasil penelitian dari parameter kecernaan bahan kering, VFA, kecernaan ADF, NDF dan hemiselulosa direkomendasikan fermentasi litter ayam selama 6 minggu.

Kata Kunci: Litter Ayam Broiler, Kandungan Kimia, Kecernaan, Fermentasi, *In vitro*

ABSTRACT

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The aim of this study was to examine effect of length of chicken litter fermentation on chemical content and *in vitro* digestibility. Completely randomized design was applied in this study with 4 treatments and 4 replications. The treatments were T0 = no fermentation; T1 = fermentation of chicken litter for 3 weeks; T2 = fermentation of chicken litter for 6 weeks; and T3 = fermentation of chicken litter for 9 weeks. Parameters observed were chemical content and digestibility value of fermented chicken litter. Different fermentation time affected the chemical content of fermented chicken litter, namely water, fat, BETN and TDN content, but did not affect ash content and fiber content. Different fermentation time affected dry matter, protein, fiber fraction digestibility (ADF, NDF, Hemicellulose), but did not affect organic matter digestibility, VFA concentration, NH_3 concentration and total protein production of chicken litter. Based on dry matter, ADF, NDF hemicellulose digestibility and VFA concentration, it is concluded that recommended ripen time for chicken litter fermentation is 6 weeks.

Key Words: Broiler Litter, Chemical Content, Digestibility, Fermentation, *In vitro*

INTRODUCTION

Poultry farming in Indonesia is becoming a widely developed as livestock industry due to the increasing demand for poultry meat by 0.3% annually (BPS, 2019). Poultry farms produce meat and eggs as the main product also produce farm waste in the form of litter. Waste from chicken farms in the form of litter consisting of husks and manure. Litter is a cage base that is generally made of husks that serve to absorb water and excreta produced by chickens during maintenance in the cage (Muharlien et al. 2011). Unprocessed litter will have a negative impact on the environment (Ibrahim & Allaily 2012). The litter usually is processed into organic fertilizer.

Chicken litter is potentially to be used as ruminant feed because it is easy to obtain and has good nutritional content. It contains 25–50% crude protein and a total digested nutrients of 55 – 60% (Rahimi et al. 2018). Manure contained in litter has a nutrient content such as crude protein by 24.9%, extract ether 2.39%, Nitrogen Free Extract 27.96%, Ca 2.31%, P 1.56% and crude fiber 23.6% (Stephenson et al. 1990). Litter has high fiber levels and there are pathogenic microorganisms in it, so it must be processed to be safe, palatabel and have high digestibility when fed to livestock. Litter processing is to eliminate pathogenic microbes, including by: chemical processing, fermentation, ensiling and heating (Bolan et al. 2010). Litter fermentation is expected to increase the

digestibility of the material so as to increase livestock productivity.

In addition to providing a more economical processing solution, litter processing is also important as an alternative way to maintain availability of feed for ruminants because of its potential nutritional content such as protein. The availability and quality of feed in the dry season tends to decrease, compared to the rainy season (Mbatha & Bakare 2018). Litter processing with fermentation can be an alternative source of feed for ruminants. Some ruminant farms have actually implemented the use of litter as animal feed. However, untreated litter can have a negative impact on livestock, due to the presence of pathogenic microorganisms in the litter. Fermented litter can be used as an option.

Fermentation is a processing material with the help of microorganisms, aiming to improve the quality of the litter, so that the complex components in the litter break down into simpler ones. Fermentation can also suppress growth of pathogenic microorganisms in the litter. Litter fermentation can reduce total number of pathogenic bacteria that can interfere with livestock health if used for feed (Najibulloh et al. 2020). The higher lactic acid bacteria population during fermentation can increase protein content of chicken litter. During the process, microorganisms will synthesize protein through the protein encryption process, and enzymes produced by microorganisms will degrade complex compounds into compounds that are easier to be digested. Fermentation can reduce crude fiber content and improve quality in terms of nutritional content and digestibility of materials due to the activity of microorganisms during fermentation (Prastyawan et al. 2012).

Previous research on chicken litter fermentation was carried out using a semi-continuous stirred reactor tank, under thermophilic conditions (55±1°C) with a fermentation duration of up to 8 months (Qiao et al. 2018), which is not applicable in the field and requires and expensive. Therefore, it is necessary to study the fermentation with an applicable and inexpensive method. This study aimed to examine chicken litter fermentation at different duration on chemical content and *in vitro* digestibility [dry matter, organic matter, protein, fiber fractions (ADF, NDF, hemicellulose), concentration of NH3 and VFA].

MATERIALS AND METHODS

The experimental design used in this study was a complete randomized design (CRD) with 4 treatments and 4 replications namely T0 = no fermentation, T1 = fermented for 3 weeks, T2 = fermented for 6 weeks, and T3 = fermented for 9 weeks. The differences among treatment means was compared using Duncan's Multiple Range Test (DMRT) at a 95% confidence level (Utama & Christiyanto 2021).

Fermentation stage

Broiler chicken litter was collected from 16 broiler cages in Cemerlang Unggas Lestari Inc, Semarang City, Central Java. The collected litter was then mixed homogeneously. The liter was weighed as much as 1 kg and then 60 grams of starter mix culture (lactic acid bacteria, cellulolytic, amylolytic and lipolytic) were added. A substrate consisting of 60 grams of mineral mix, 60 grams of salt, 60 ml of molasses mixed with 100 ml of water, were added to the litter. All ingredients are mixed homogeneously, and put into a fermentation plastic container that is tightly tied. The litter was then fermented according to the treatment (T0, T1, T2 and T3). After the fermentation was complete, all samples were dried in a mesh-covered tray to avoid contamination by microorganisms under the sun for 12 hours. The dried samples were ground using a blender until they were powdered and ready for analysis.

Chemical content analysis

Proximate analysis consisting of moisture content, ash content, crude fat, crude protein and crude fiber was carried out using the AOAC method (2005). The content of nitrogen free extract is calculated using the formula according to the method of (Pratiwi et al. 2015), while the Total Digestible Nutrients (TDN) is calculated using equation according (Widodo et al. 2012).

Nitrogen Free Extract:

$$100 - (\text{ash} + \text{crude fat} + \text{crrude fiber} + \text{crude Protein}$$

$$TDN = 70{,}6 + 0{,}259a + 1{,}01b - 0{,}76c + 0{,}0991d$$

where a is percentage of digestible crude protein, b is percentage of crude fat, c is percentage of crude fiber, and d is percentage of NE.

In vitro digestibility

In vitro parameter measurements were carried out by making conditions in accordance to the actual rumen, the experiment was carried out using the Tilley & Terry (1963) method. This technique uses an artificial rumen in the form of a 100 ml fermenter tube, McDougall's solution as a substitute for saliva and fresh cow rumen from the Slaughterhouse as a source of inoculum. The sample was put as much as 0.56 g into a sterilized fermenter tube, then the sample was given 40 ml of McDougall's solution and 10 ml of beef rumen fluid. The fermenter tube is then added with CO_2 gas for 10 – 20 seconds to create anaerobic conditions and closed with a rubber cap. The tubes were then incubated

in a water bath for 3 hours and after incubation the tubes were placed in ice water to stop the fermentation. Each parameter was tested using 32 tubes for *in vitro*.

Meanwhile, the measurement of NH3 and VFA concentrations was carried out using the supernatant. The supernatant used came from the results of the first incubation for 3 hours (before the addition of HCl).

Dry matter and organic matter digestibility

Digestibility of dry matter and organic matter was calculated using the formula:

$$\textit{In vitro}\ \text{Dry Matter digestibility} = \frac{\text{A - (B-C)}}{\text{D}} \text{ x 100}$$

where A is dry matter weight of the sample in gram, B is residual dry matter weight in gram, C is blank dry matter weight in gram, and D is dry matter weight of the sample in gram.

$$\textit{In vitro}\ \text{organic matter digestility} = \frac{\text{E - F - G}}{\text{H}}\ x\ 100$$

where E is organic matter weight of the sample in gram, F is residual organic matter weight in gram, G is blank organic matter weight in gram, and H is dry matter weight of the sample in gram.

Total protein

The sample was mixed with 40 ml of McDougall's solution and 10 ml of rumen fluid in the fermenter tube which was then folowed with CO_2 gas. The incubation was carried out for 48 hours at a temperature of 39^0C. After incubation, 10 ml of the sample was taken and TCA + SSA was added to settle for 4-5 hours and filtered. The filtered residue was analyzed for protein by the Kjeldhal method.

Measurement of *in vitro* total protein used Kjeldahl method. Sample proteins and residual proteins were analyzed by Kjeldahl method according to AOAC, (2005). Total protein can be calculated using the formula (Sumadi et al. 2017):

$$\text{Total Protein} = \frac{\text{(I-J)}\times\text{K}\times 14\times 6{,}25}{L}\ x\ mg/g$$

where I is ml HCl titrant, J is ml HCl blank, K is N HCl, L is Sample weight of sediment (mg/g) of residue.

Protein digestibility

Protein digestibility was calculated accoding to kjeldahl method (1963). Sample proteins and residual proteins were analyzed by kjeldahl method according to AOAC (2005). The formula of protein digestibility calculation is as follows:

$$\text{Protein Digestibility} = \frac{\text{M}\times\text{N - P}}{\text{M}\times\text{N}}\ x\ 100\%$$

where M is % crude protein sample, N is weight of samples, P is % crude protein content of residue x weight of residue.

Concentration of volatile fatty acid (VFA)

The VFA total (Mm) concentrations were determined by steam distillation method (General Laboratory procedure) (Abbaticehlo et al. 1983) and calculating as:

$$\text{Blank titrant volume} - \text{sample titrant volume}) \times \text{N} - \text{HCL} \times \frac{1000}{5})$$

where N-HCL is Normality of HCL, Blank titrant volume is Number of HCL titer for 5 ml NaOH (blank), and sample titrant volume is Number of HCL titer to dissolve the distillate.

Ammonia concentration (NH_3)

Analysis of ammonia concentration (NH_3) using spectrophotometer method (Azizah & Humairoh 2015).

Digestibility of NDF, ADF and hemicellulose

The method used in this study was an experimental method, namely the analysis of the digestibility of ADF, NDF, Lignin and Hemicellulose by *in vitro*. Fiber analysis used Van Soest method, while calculation of values calculated based on Tilley & Terry (1963).

$$\%\text{Digestibility of NDF:}\quad \frac{\text{NDF sample (g)-(NDF residue (g)-NDF Blanko (g))}}{\text{NDF sample (g)}}\ x\ 100\%$$

$$\%\ \text{Digestibility of ADF:}\quad \frac{\text{ADF sample (g)-(ADF residue (g)-ADF Blanko (g))}}{\text{ADF sample (g)}}\ x\ 100\%$$

$$\%\ \text{Hemicellulose Digestibility:}\quad \frac{\text{hemicel. sample (g)-(hemicel. residue (g)-hemicel. blanko (g))}}{\text{hemicel. sample (g)}}\ x\ 100\%$$

where hemicel. is hemicellulose.

RESULTS AND DISCUSSION

Analysis of variance showed that T3 was significantly different from T0, T1, T2 ($P<0.05$), and T1 was not significantly different from T2 on the water content of fermented litter. Treatment T3 had a higher water content than treatments T0, T1, and T2. Factors that cause an increase in water content can be caused by microorganisms using dry matter substrates for development and growth during the fermentation process, causing a decrease in dry matter levels and resulting in an increase in fermented water content

(Driehuis et al. 1997). The average water content was 44.09%. Litter has an average moisture content ranging from 16,32 - 19,14% (Marang et al. 2019). The increase occurred in the 63rd day fermentation. The increase in water content can also be caused by the fermentation process. During the process, there is a decrease in dry matter and an increase in water content caused by the first fermentation stage, namely during respiration, glucose is converted into CO_2, H_2O and heat (McDonald 1981).

Analysis of variance showed that difference in fermentation time was not significant (P>0.05) on ash content. Different ripen time had no significant effect in reducing fermented litter ash content. This was because the starter used organic matter as a source of nutrition and at the same time breaks down crude fiber into simple carbohydrates so that organic matter increases. The process will affect the content of organic matter because the compound will degrade complex compounds into simple ones (Setyawati et al. 2014). The average ash content in the litter was 33%, this value was higher than the results reported by Chaudhry et al. (1993) (17.8%). The ash content increased starting from day 0 to day 63. An increase in ash content of fermented litter occurs because during fermentation process there is a decrease in organic matter due to substrate degradation by starter microbes (Collett 2012).

Analysis of variance showed that T0 was significantly different from T1, T2, T3 (P<0.05) on the crude protein content of fermented litter. In fermented litter there is a decrease in value of crude protein. Fermentation is carried out with the aim of increasing the nutritional value of feed ingredients, especially at increasing protein levels (Prastyawan et al. 2012). The protein value of the litter was reported 18.9% (Chaudhry et al. 1993). The decrease in crude protein levels started from T0 treatment to T2 treatment which was caused by the number of mixed microorganisms in the feed being less than optimal. The increase in protein content occurred in treatment T3 on day 63 into 19.27%. Factors that cause an increase in protein content in the activity of microorganisms that hydrolyze proteins in the substrate. Microorganisms will hydrolyze proteins in the substrate with the help of proteolytic enzymes produced by lactic acid bacteria (Hilakore et al. 2013).

Analysis of variance showed that T0 was significantly different from T1, T2 (P<0.05) on crude fat content of fermented litter. There was a decrease in the crude fat content among the treatment. The decrease is influenced by the process due to the reshuffle of cell wall composition and saponification reaction so that water-soluble cell walls become dissolved. Average crude fat content in the litter is 2.2%. Crude fat content in chicken litter is 1.22% (Setyaningrum & Ismail 2019). Litter fermentation process has microbial activity that produces high fatty acids so that the fat content increases (Bakshi & Fontenot 1998). The increased free fat content is utilized by lipolytic microorganisms as an energy source, resulting in a decrease in crude fat content (Suningsih et al. 2019).

Analysis of variance showed that effect of the treatment was not significantly different (P>0.05) on crude fiber content of fermented litter. The average crude fiber was 19.62%. Crude fiber content in fermented litter is 13.24% (Telew et al. 2013). The factor causing no decrease in fiber content is the high crude fiber content of components that make up the litter, especially lignin and cellulose. The litter used is derived from husk material. Microorganisms during the fermentation process are difficult to degrade lignin (Ratnakomala et al. 2006). The high content of lignin and cellulose in the litter decreased the ability of microorganism enzymes to digest crude fiber content of the feed. The value of fiber content is still relatively safe to be given to ruminants. Digestion of crude fiber in ruminants occurs in the rumen with the help of microorganisms (Irawati et al. 2019).

Analysis of variance showed that T0 was significantly different from T1, T2, T3 (P<0.05) and T2 was not significantly different from T3 on the content of nitrogen free extract. Compared to control, NFE increased due to fermentation (T1, T2 and T3). The highest increased of NFE content was in T1. The increased of NFE content due to the increased of bacteria population so that they degrade complex compounds into simple compounds. The decrease in fiber content in feed ingredients will increase the NFE content (Pratiwi et al. 2015).

Analysis of variance showed that different length of fermentation affected TDN content. T0, T3 was significantly contained higher TDN than T1, T2 (P<0.05). Litter of organic chicken has a TDN content of 55-60% (Kwak et al. 2008). The increase in TDN levels occurred due to a decrease in crude fiber in T3 treatment by the cellulose enzyme produced by starter microorganisms, thereby increasing digestibility of feed nutrients (Amrullah 2019). The higher the value the better the quality of the feed, this is due to the more nutrients being digested by the animal's body (Riyanto et al. 2020). Digestibility of dry matter, organic matter, Volatile Fatty Acids (VFA), ammonia (NH_3), crude protein digestibility of the fermented litter is presented in Table 2.

Dry Matter Digestibility

In vitro DM digestibility of fermented chicken litter in sheep rumen was influenced by different fermentation times (P<0.05). Dry matter digestibility of T0 was significantly different from T1 and T2, but not significantly different from T3, the DM digestibility of T2 was the highest. The value of this research results is

Table 1. Water content, ash, crude fat, crude protein, crude fiber, BETN and TDN of fermented chicken litter at different fermentation times

Parameter	Fermentation Duration			
	T0	T1	T2	T3
Water Content (%)	38.39 ± 1.02 [c]	43.34 ± 1.31 [b]	44.30 ± 1.08 [b]	50.34 ± 1.59 [a]
Ash (%)	31.26 ± 0.43	31.38 ± 2.53	32.30 ± 0.79	33.08 ± 1.57
Crude Protein (%)	25.73 ± 1.06 [a]	18.34 ± 1.10 [b]	18.12 ± 0.52 [b]	19.27 ± 0.71 [b]
Crude Fat (%)	2.69± 0.36 [a]	1.85 ± 0.65 [b]	1.79 ± 0.39 [b]	2.50 ± 0.38 [ab]
Crude Fiber (%)	19.52± 1.08	19.90 ± 1.13	20.83 ± 1.58	18.22 ± 1.82
NFE (%)	20.80 ± 1.26 [c]	28.53 ± 1.46 [a]	26.96 ± 0.43 [b]	26.93 ± 1.97 [b]
TDN (%)	48.57 ± 2.15 [a]	44.49 ± 3.66 [b]	42.62 ± 1.73 [b]	45.92 ± 1.90 [a]
ADF Level (%)	26.17±0.40 [c]	30.91±0.76 [a]	28.60±0.16 [b]	31.80±0.93 [a]
NDF Level (%)	40.11±0.54 [a]	37.91±0.44 [b]	36.60±0.35 [c]	34.32±0.57 [d]

NFE = Nitrogen free extract, TDN = Total digestible nutrient, ADF= Acid detergent fiber, NDF= Neutral detergent fiber. Different superscripts on the same line indicate significant differences (P <0.05)

Table 2. Dry matter digestibility, organic matter digestibility, VFA, NH_3, and Crude protein digestibility, total protein production of fermented chicken litter at different fermentation times

Parameter	Fermentation Duration			
	T0	T1	T2	T3
Dry matter digestibility (%)	48.4±0.46 [c]	51.5±0.56 [b]	54.8±0.63 [a]	48.9±0.69 [c]
Organic matter digestibility (%)	71.9±1.79	69.8±1.39	70.1±1.22	68.9±1.28
VFA (mM)	77.5±5.00	80.0±5.77	85.0±11.55	75.0±11.55
NH_3 N (mM)	25.5±2.50	27.3±5.85	24.2±7.29	26.3±2.77
Total Protein (mg/g)	1632±140	1373±145	1458±114	1180±129
Crude protein digestibility (%)	43.6±0.58 [b]	43.7±0.74 [b]	45.2±0.98 [a]	45.8±0.48 [a]

Mean values within a row with different superscripts differ significantly (P≤0.05)

almost the same as that reported by Jokthan et al. (2013) which stated that the dry matter digestibility litter value was 44.31 – 51.33%. The lower dry matter digestibility value in T0 could occur because there was no time for fermentation in that treatment, so the components of the litter at T0 were more complex. The more complex litter components at T0 made the degrading bacteria *in vitro* not optimally degrade the litter, thus giving the lowest dry matter digestibility yield.

The low digestibility value was affected by microbial activity of the rumen fluid, content of feed ingredients used, and type of feed. Priyanto et al. (2017) stated that the factors that influence dry matter digestibility value can come from: form of feed, composition in feed, and microbial activity in the rumen fluid. The low dry matter digestibility value also occurs due to the low ability of rumen microorganisms to digest litter components. Setiyaningsih (2013) stated that the low dry matter digestibility value could be caused by microbial conditions in rumen fluid that could not utilize nutritional content of feed ingredients. Components that are difficult to digest are especially crude fiber composition of the litter that comes from husks. Krogdahl & Dalsgard (1981) stated that the digestibility of feed ingredients is influenced by several factors including form of feed, feed composition and nutritional content of feed but not affected by pathogenic bacteria *Escherichia coli* and *Staphylococcus aureus*, that present in chicken litter.

Organic matter digestibility

In vitro digestibility of fermented chicken litter organic matter with sheep rumen was not affected by different ripen times (P>0.05). The digestibility value ranged from 69.8 - 71.9%. The digestibility value in this study was higher than the results reported by Hadjipanayiotou (1982), litter organic matter in this study indicated that either fermented or not fermented chicken litter h the potential to be used as an alternative feed for ruminants. Al-Arif et al. (2017) stated that the fermentation treatment resulted in a

degradation process of fiber fractions such as cellulose, hemicellulose and lignin and has an impact on increasing the digestibility value. The digestibility of organic matter can be influenced by the digestibility of components of organic matter, namely protein, fat and carbohydrates.

The digestibility value can be influenced by nutrition content of the material especially fiber and ash content. The relatively similar organic matter digestibility values were thought to be due to the same fiber content and fermented litter ash content (Table 2). Suharlina et al. (2017) stated that digestibility value of organic matter in feed ingredients is determined by nutrition content present in the material, especially fiber, that will be difficult to digest by rumen microbes. The ash content of fermented chicken litter was not affected by the treatment, so that the digestibility of organic matter was not too influential. This is because organic matter is all the nutrients in the fermented chicken litter except the ash. Al-Arif et al. (2017) stated that digestibility value of organic matter can be used to measure the total amount of nutrients that can be absorbed in the digestive tract of ruminants, estimate protein synthesis of microorganisms and measure the energy produced, where dry matter digestibility is affected by the presence of ash content.

Concentration of volatile fatty acid (VFA)

Volatile Fatty Acid (VFA) *in vitro* of fermented chicken litter was not affected by different fermentation times (P>0.05). The concentration ranged from 75 - 77.5 Mm. Sutardi (1997) stated that good rumen microbial VFA concentrations ranged from 80 – 160 Mm. The VFA measured in this study was the total VFA, namely propionate, acetate and butyrate. The value was still below the good VFA standard for ruminants, indicated that fermented chicken litter can't be used as a single feed, but must be combined with other feed ingredients, so that VFA concentrations can be achieved according to the standard. VFA that is too low will inhibit activity of rumen microorganisms. Singh et al. (2020) stated that in addition to being influenced by the substrate used, activity of microorganisms was also influenced the VFA concentration.

Ripen time did not affect VFA concentration, presumably because crude fiber content in fermented chicken litter was also relatively the same. Widodo & Sutrisno (2012) stated that the fiber fraction in feed will be converted into simple sugars which undergo glycolysis to pyruvic acid and become VFA. The amount of soluble carbohydrates in the fermented chicken litter was assumed to be the same, so the VFA produced was also the same. Wijayanti et al. (2012) stated that factors that affect concentration of VFA include: amount of feed, feed fermentability, amount of soluble carbohydrates, type and pH of the rumen.

Concentration of ammonia (NH_3) in sheep

Concentration of NH_3 in fermented chicken litter *in vitro* was not affected by different ripen times (P>0.05). The NH_3 concentration ranged from 24.2 - 27.3 mM. This value is higher than that reported by Sandi et al. (2016) which states that the levels of NH_3 to support the growth of rumen microbes are in the range of 6-21 mM. Cabeza et al. (2018) stated ammonia is the result of degradation of protein and non-protein nitrogen (NPN) that enters the rumen of ruminants.

Different fermentation time did not affect the production of NH_3 which could be due to relatively the same protein degradation process in the fermented litter. Wijayanti et al. (2012) stated that the factors that affect the concentration of NH_3 are carbohydrates in the ration of the amount of feed, protein degradation, as well as solubility and rumen pH. The higher NH_3 concentration of fermented chicken litter indicated that more protein was hydrolyzed into ammonia. Prayitno et al. (2018) stated that a high NH_3 value is influenced by level of protein solubility in the feed, the higher the protein solubility in the feed, the more easily the protein will be degraded by microbes. NH_3 for rumen microorganisms acts as the main nitrogen source to support the protein synthesis process.

Total protein

Total protein of fermented chicken litter *in vitro* was not affected by different fermentation times (P>0.05). The value was 1180.167 – 1632.49 mg/g. Pal et al. (2016) stated that several factors that affect total protein include NH_3 production, carbon skeletons, and energy sources. Total protein is an indication of microbial protein from the rumen and protein litter of fermented chicken that is not degraded in the rumen of ruminants. Sumadi et al. (2017) stated that total protein plays a role in evaluating the value of protein that escapes degradation of rumen microorganisms, as well as how much concentration of microbial protein is in the post-rumen digestive organs. Priyanto et al. (2017) stated that high total protein can occur due to ideal conditions and the availability of energy sources as quickly as the formation of NH_3, so that when NH_3 is formed, the fermentation product from carbohydrates will function as an energy source and carbon source.

Crude protein digestibility

In vitro crude protein digestibility in this study was influenced by different fermentation times (P<0.05). Compared to control, crude protein digestibility of T2

Table 3. Digestibility of fermented chicken litter fiber fraction at different ripening time

Parameter	Fermentation Duration			
	T0	T1	T2	T3
ADF digestibility (%)	35.3±0.31 [d]	47.9±0.93 [b]	49.4±0.57 [a]	44.5±0.97 [c]
NDF digestibility (%)	45.9±1.00 [d]	59.4±0.51 [b]	64.1±0.85 [a]	54.9±1.10 [c]
Hemicellulose digestibility (%)	20.8±0.96 [b]	17.3±0.66 [a]	21.9±1.03 [b]	15.1±0.57 [a]

Different superscripts on the same line indicate significant differences (P <0.05)

and T3 were higher, but not significantly different from T1. The high values in T2 and T3 could be due to the crude protein and TDN content in the fermented chicken litter which was more optimally utilized compared to other fermented chicken litters. Teti et al. (2018) stated that high crude protein content in the ration will increase the rate of rumen microbial population and the ability to degrade feed increases, besides being influenced by crude protein and TDN levels.

The highest protein content of fermented chicken litter was at T0, but not comparable to the digestibility value of the protein produced, thought to occur due to chemical processes during fermentation and protein was not digested optimally by microorganisms *in vitro*. Ayasan et al. (2018) stated that the increase in protein digestibility was related to the degradation of trypsin inhibitors and the process of nucleic acid loss of secondary and tertiary structures (protein denaturation). T2 and T3 treatments gave the best crude protein digestibility value because the N content was proportional to the TDN content in the material, due to the optimal fermentation time. Ayuningsih et al. (2018) stated that the supply of nitrogen balanced with high TDN will lead to a balance of protein and energy which has an impact on digestibility and higher feed efficiency.

Fiber digestibility

The digestibility of ADF, NDF, hemicellulose of fermented chicken litter at different ripening times is presented in Table 3.

Acid detergent fiber (ADF) digestibility

Digestibility of ADF of fermented chicken litter was influenced by different fermentation times (P<0.05). Rahimi et al. (2018) stated that the digestibility value of ADF in various types of broiler litter was 38.11–43.20%. This value indicates that there is an effect of the length of fermentation to increase the digestibility of ADF. Hambakodu et al. (2020) stated that the digestibility value of ADF is a combination of digestibility value which contains cellulose, and lignin. Different fermentation times affect the levels of ADF thus affecting better acceptance of rumen microorganisms or affecting ADF digestibility. Wijaya et al. (2018) stated that digestibility of ADF in sheep was caused by adaptation of rumen microbes to a feed ingredient. The highest value of ADF digestibility was in T2. This shows that ripen time for 6 weeks increases the digestibility of ADF in broiler litter. The increase was caused by the breaking of the lignin-cellulose bonds by microbes during fermentation. Putri (2020) stated that fermentation causes lignin cellulose bonds consisting of lignin, cellulose and also hemicellulose to be broken so that they are easily digested by rumen microbes.

Neutral Detergent Fiber (NDF) Digestibility

NDF digestibility of fermented chicken litter was influenced by different fermentation times (P<0.05). All treatments were significantly different from each other. NDF is the main component of fiber in fermented chicken litter. The presence of NDF in chicken litter comes from the husk which is part of the litter which is high in fiber content. Zhao et al. (2019) stated that NDF is a constituent of cell walls consisting of hemicellulose, lignin, cellulose, and other small components such as silicates and proteins. Turangan et al. (2018) stated that NDF digestibility levels were influenced by crude fiber content such as lignin, silica, energy sources, protein, minerals and vitamins. In fermentation process there are enzymes that work in the process of breaking lignin bonds from fermented chicken manure. Yanti et al. (2021) stated that the penetration of rumen microorganism enzymeswould be easier to degrade NDF due to the presence of lignase enzymes that break the bonds of lignohemicellulose and lignocellulose.

Hemicellulose digestibility

Hemicellulose digestibility of fermented chicken litter was influenced by different length of fermentation (P<0.05). Treatment T0 was significantly higher than T1 and T3, but not significantly different from T2; T1 was significantly different from treatment T2 and T3; T2 and T3 were significantly different. The digestibility

of the material is influenced by crude fiber content, because crude fiber content will result in a low degradation value. Angelidaki & Ahring (2000) stated that crude fiber in the form of cellulose and hemicellulose often binds to lignin, and will be difficult to be broken down by digestive enzymes, which causes lower digestibility if a feed ingredient contains high fiber.

Higher hemicellulose digestibility results at T2 can occur because in this treatment digestibility of NDF is also high, so it has an impact on high hemicellulose digestibility as well. Yanti et al. (2021) stated that digestibility of hemicellulose is generally higher than digestibility of cellulose and digestibility value is influenced by levels of NDF and ADF. The hemicellulose constituent fractions are generally more easily digested by rumen microorganisms. Zhao et al. (2019) stated that hemicellulose has an amorphous structure and low polymerization rate, so it will be easier to digest than other cell wall components. The use of cellulose and hemicellulose as a source of carbohydrates will be easily fermented by rumen bacteria into VFA which is a source of energy for the growth of sheep.

CONCLUSION

Fermentation increased water and NFE content, decreased CP, Crude Fat, NDF and TDN, but did not affect ash and CF content. Fermentation increased dry matter digestibility, protein digestibility, fiber fraction digestibility (ADF, NDF, Hemicellulose). The recommended treatment is fermenting chicken litter for 6 weeks.

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REFERENCES

Abbaticehlo P, Bailee V, Cera O. 1983. Determination of volatile fatty acids by gas chromatography after a rapid steam distillation of wastes. Environ Technol Lett. 4:179–182. DOI:10.1080/09593338309384191.

Al-Arif MA, Suwanti LT, Estoepangestie AS, Lamid M. 2017. The nutrients contents, dry matter digestibility, organic matter digestibility, total digestible nutrient, and NH_3 rumen production of three kinds of cattle feeding models. KnE Life Sci. 3:338. DOI:10.18502/kls.v3i6.1142.

Amrullah M, Tampoebolon BIM, Prasetyono BW. 2019 Kajian pengaruh proses fermentasi sekam padi amoniasi menggunakan aspergillus niger terhadap serat kasar, protein kasar, dan total digestible nutrients. JPPP. 16:25–31.

Angelidaki I, Ahring BK. 2000. Methods for increasing the biogas potential from the recalcitrant organic matter contained in manure. Water Sci Technol. 41(3):189–194. DOI: 10.2166/wst.2000.0071.

Ayasan T, Ülger I, Kaliber M, Ergül S, Inci H, Mart D, Türkeri M. 2018. Comparison of *in vitro* gas production, nutritive value, metabolizable energy and organic matter digestibility of some chickpea varieties. Iran J Appl Anim Sci. 8:31–136.

Ayuningsih B, Hernaman I, Ramdani D, Siswoyo S. 2018. Pengaruh imbangan protein dan energi terhadap efisiensi penggunaan ransum pada domba Garut betina. J Ilm Peternak Terpadu. 6:97. DOI:10.23960/jipt.v6i1.p97-100.

Azizah M, Azizah M. 2015. Analisis kadar amonia (NH3) dalam air sungai Cileungsi. Nusa Sylva. 15:47–54.

Bakshi MPS, Fontenot JP. 1998. Processing and nutritive evaluation of broiler litter as livestock feed. Anim Feed Sci Technol. 74:337–345. DOI:10.1016/S0377-8401(98)00181-3.

Bolan NS, Szogi AA, Chuasavathi T, Seshadri B, Rothrock MJ, Panneerselvam P. 2010. Uses and management of poultry litter. Worlds Poult Sci J. 66:673–698. DOI: 10.1017/S0043933910000656.

[BPS] Badan Pusat Statistik. 2019. Broiler Population by Province. Jakarta (Indones): Bada Pusat Statistik.

Cabeza I, Waterhouse T, Sohi S, Rooke JA. 2018. Effect of biochar produced from different biomass sources and at different process temperatures on methane production and ammonia concentrations *in vitro*. Anim Feed Sci Technol. 237:1–7. DOI:10.1016/j.anifeedsci.2018.01.003.

Chaudhry SM, Naseer Z, Chaudhry DM. 1993. Fermentation characteristics and nutritive value of broiler litter ensiled with corn forage. Food Chem. 48:51–55. DOI: 10.1016/0308-8146(93)90220-A.

Collett SR. 2012. Nutrition and wet litter problems in poultry. Anim Feed Sci Technol. 173:65–75. DOI:10.1016/j.anifeedsci.2011.12.013.

Driehuis F, Van Wikselaar PG, Van Vuuren AM, Spoelstra SF. 1997. Effect of a bacterial inoculant on rate of fermentation and chemical composition of high dry matter grass silages. J Agric Sci. 128:323–329. DOI: 10.1017/S0021859696004157.

Hadjipanayiotou M. 1982. Laboratory evaluation of ensiled poultry litter. Anim Prod. 35:157–161. DOI: 10.1017/S0003356100000921.

Hambakodu M, Kaka A, Ina YT. 2020. Kajian In Vitro Kecernaan Fraksi Serat Hijauan Tropis pada Media Cairan Rumen Kambing. JITPT. 7:29. DOI:10.33772/jitro.v7i1.8907.

Ibrahim S, Allaily A. 2012. Pengaruh berbagai bahan litter terhadap konsentrasi ammonia udara ambient kandang dan performan ayam broiler. J Agripet. 12:47–51. DOI: 10.17969/agripet.v12i1.288.

Irawati E, Purnamasari E, Arsyad F. 2019. Kualitas fisik dan nutrisi eceng gondok (*Eichornia crassipes*) dengan lama fermentasi yang berbeda. J Peternak. 16:18. DOI:10.24014/jupet.v16i1.3679.

Jokthan GE, Muhammad SA, Osuhur CU. 2013. Effect of Cottonseed cake replacement with broiler litter on performance of Yankasa Rams fed maize husk basal diets. 2013:147–155.

Krogdahl A, Dalsgard B. 1981. Estimation of nitrogen digestibility in poultry: content and distribution of major urinary nitrogen compounds in excreta. Poult Sci. 60:2480–2485. DOI:10.3382/ps.0602480.

Kwak WS, Jung SH, Kim YI. 2008. Broiler litter supplementation improves storage and feed-nutritional value of sawdust-based spent mushroom substrate. Bioresour Technol. 99:2947–2955. DOI: 10.1016/j.biortech.2007.06.021.

Marang EAF, Mahfudz LD, Sarjana TA, Setyaningrum S. 2019. Kualitas dan kadar amonia litter akibat penambahan sinbiotik dalam ransum ayam broiler. J Peternak Indones. 21:303. DOI:10.25077/jpi.21.3.303-310.2019.

Maritje Aleonor Hilakore, Suryahadi, Komang Wiryawan DM. 2013. Peningkatan Kadar Protein Putak melalui Fermentasi oleh Kapang *Trichoderma reesei*. 14:250–254.

Mbatha KR, Bakare AG. 2018. Browse silage as potential feed for captive wild ungulates in southern Africa: A review. Anim Nutr. 4:1–10. DOI:10.1016/j.aninu.2017.12.003.

McDonald P. 1981. The Biochemistry of Silage. London: John Wiley and Sons Ltd.

Muharlien, Achmanu, R.Rachmawati. 2011. Meningkatkan Produksi Ayam Pedaging Melalui Pengaturan Proporsi Sekam, Pasir Dan Kapur Sebagai Litter. J Ternak Trop. 12:38–45.

Najibulloh M, Ulupi N, Salundik S. 2020. Pengaruh daur ulang litter terhadap kualitas litter dan udara dalam pemeliharaan broiler. Livest Anim Res. 18:107. DOI: 10.20961/lar.v18i2.42932.

Nur Eka Styawati, Muhtarudin L. 2014. Pengaruh lama fermentasi trametes sp. Terhadap kadar bahan kering, kadar abu, dan kadar serat kasar daun nenas varietas smooth cayenne. J Ilm Peternak Terpadu. 2:19–24.

Pal RS, Singh KS, Tripathi MK. 2016. Nutrient intake, digestibility, milk production and composition of lactating cows fed oat hay and concentrate containing varying levels of poultry litter. Indian J Anim Res. 50:194–198. DOI:10.18805/ijar.9540.

Prastyawan RMP, Tampoebolon BIM, Surono. 2012. Peningkatan kualitas tongkol jagung melalui teknologi amoniasi fermentasi (amofer) terhadap kecernaan bahan kering dan bahan organik serta protein total secara in vitro. Anim Agric J. 1:611–621.

Pratiwi I, Fathul F, Muhtarudin. 2015. The effect of different additioning starter to making silage on crude fiber content, crude fat, water content, and material extract without nitrogen silage. J Ilm Perten Terpadu. 3:116–120.

Prayitno RS, Wahyono F, Pangestu E. 2018. Pengaruh suplementasi sumber protein hijauan leguminosa terhadap produksi amonia dan protein total ruminal secara in vitro. J Peternak Indones. 20:116. DOI: 10.25077/jpi.20.2.116-123.2018.

Priyanto A, Endraswati A, Rizkiyanshah, Febriyani NC, Nopiansyah T, Nuswantara LK. 2017. Pengaruh pemberian minyak jagung dan suplementasi urea pada ransum terhadap profil cairan rumen (KcBK, KcBO, pH, N-NH 3 dan Total Mikroba Rumen). J Ilmu Ternak. 17:1–9.

Putri PW. 2020. Kandungan neutral detergent fibre (NDF), acid detergent fibre (ADF), hemiselulisa, selulosa dan lignin onggok yang difermentasi trichoderma reesei dengan suplementasi N, S, P. Bull Appl Anim Res. 2:33–37. DOI:10.36423/baar.v2i1.227.

Rahimi MR, Alijoo YA, Pirmohammadi R, Alimirzaei M. 2018. Effects of feeding with broiler litter in pellet-form diet on qizil fattening lambs' performance, nutrient digestibility, blood metabolites and husbandry economics. Vet Res Forum. 9:245–251. DOI:10.30466/vrf.2018.32081.

Singh R, Park S, Koo JS, Balasubramanian B. 2020. Influence of various concentrations of aflatoxin B 1 on in vitro rumen fermentation of a buffalo diet. Korean J Agric Sci. 47:131–138. DOI:10.7744/kjoas.20200005.

Ratnakomala S, Ridwan R, Kartina G, Widyastuti Y. 2006. The effect of *Lactobacillus plantarum* 1A-2 and 1BL-2 inoculant on the quality of napier grass silage. Biodiversitas J Biol Divers. 7:131–134. DOI:10.13057/biodiv/d070208.

Riyanto J, Widyawati SD, Sudibya S. 2020. Pengaruh perbedaan rasio menir kedelai proteksi dan tanpa proteksi terhadap konsumsi, kecernaan dan nilai nutrien pakan Domba Ekor Gemuk. Livest Anim Res. 18:240. DOI:10.20961/lar.v18i3.45995.

Sandi S, Ali AIM, Akbar AA. 2016. Uji *in-vitro* wafer ransum komplit dengan bahan perekat yang berbeda. J Peternak Sriwij. 4:7–16. DOI:10.33230/jps.4.2.2015.2802.

Setiyaningsih KDMC dan S. 2013. Kecernaan bahan Kering dan bahan organik secara in vitro hijauan Desmodium cinereum pada berbagai dosis pupuk organik cair dan jarak tanam. J Chem Inf Model. 53:1689–1699.

Setyaningrum S, Ismail D. 2019. Efektivitas pupuk kandang dari kotoran sapi, domba dan ayam terhadap kadar lemak kasar, protein kasar dan serat kasar rumput gajah pada defoliasi kedua. J Anim Sci Agron. 4:18–28.

Stephenson AH, McCaskey TA, Ruffin BG. 1990. A survey of broiler litter composition and potential value as a nutrient resource. Biol Wastes. 34:1–9. DOI: 10.1016/0269-7483(90)90139-J.

Suharlina S, Abdullah L, Astuti D, Nahrowi N, Jayanegara A. 2017. Karakteristik fermentasi rumen terhadap beberapa jenis tanaman leguminosa. Pastura. 5:71. DOI: 10.24843/pastura.2016.v05.i02.p02.

Sumadi S, Subrata A, Sutrisno S. 2017. Produksi Protein total dan kecernaan protein daun kelor secara in vitro. J Sain Peternak Indones. 12:419–423. DOI: 10.31186/jspi. id.12.4.419-423.

Suningsih N, Ibrahim W, Liandris O, Yulianti R. 2019. Kualitas fisik dan nutrisi jerami padi fermentasi pada berbagai penambahan starter. J Sain Peternak Indones. 14:191–200. DOI:10.31186/jspi.id.14.2.191-200.

Sutardi T. 1997. Peluang dan tantangan pengembangan ilmu-ilmu nutrisi ternak. Bogor (Indones): IPB University.

Telew C, Kereh VG, Untu IM, Rembet B. 2017. Pengayaan nilai nutritif sekam padi berbasis bioteknologi “effective microorganisms” (EM4) sebagai bahan pakan organik. Zootec. 32:1–8. DOI:10.35792/zot.32.5.2013.983.

Teti N, Hernaman I, Ayuningsih B, Ramdani D, Siswoyo 2018. Pengaruh imbangan protein dan energi terhadap kecernaan nutrien ransum domba garut betina. J Ilmu Teknol Peternak. 2:97–101.

Tilley JMA, Terry RA. 1963. A Two-stage technique for the *in vitro* digestion of forage crops. Grass Forage Sci. 18:104–111. DOI:10.1111/j.1365-2494.1963.tb00335.x.

Turangan, G. G., B. Tulung, Y. R. L. Tulung MRW. 2018. Kecernaan NDF dan ADF yang mendapat suplementasi urea molasses multinutrient block (UMMB) dari beberapa jenis limbah pertanian dan rumput lapang pada sapi Peranakan Ongole (PO). Zootec. 38:320–328.

Utama CS, Christiyanto M. 2021. The feasibility of fermented litter as a feed ingredient for ruminant livestock. J Adv Vet Anim Res. 8:312–322. DOI:10.5455/javar.2021 .h517.

Wei Q, ShaoJie B, LinPeng X, Ren, ZhengRan R, Renjie D. 2018. Effect of feed concentration on long-term thermophilic methane fermentation of chicken manure. China Environ Sci. 38:2593–2601.

Widodo, F W, Sutrisno. 2012. Kecernaan Bahan Kering, Kecernaan Bahan Organik, Produksi Vfa Dan Nh3 Pakan Komplit Dengan Level Jerami Padi Berbeda Secara in Vitro. Anim Agric J. 1(1):215–230.

Wijaya AS, Dhalika T, Nurachma S. 2018. Pengaruh pemberian silase campuran Indigofera sp. dan rumput gajah pada berbagai rasio terhadap kecernaan serat kasar dan BETN pada domba garut jantan. J Ilmu Ternak Univ Padjadjaran. 18:51. DOI:10.24198 /jit.v18i1.16499.

Wijayanti E, Wahyono F, Surono. 2012. Kecernaan nutrien dan fermentabilitas pakan komplit dengan level ampas tebu yang berbeda secara *in vitro*. Anim Agric J. 1:167–179.

Yanti G, Jamarun N, Elihasridas, Astuti T. 2021. Quality improvement of sugarcane top as animal feed with biodelignification by phanerochaete chrysosporium fungi on *in-vitro* digestibility of NDF, ADF, Cellulose and hemicellulose. J Phys Conf Ser. 1940. DOI: 10. 1088/1742-6596/1940/1/012063.

Zhao J, Dong Z, Li J, Chen L, Bai Y, Jia Y, Shao T. 2019. Effects of sugar sources and doses on fermentation dynamics, carbohydrates changes, *in vitro* digestibility and gas production of rice straw silage. Ital J Anim Sci. 18:1345–1355. DOI:10.1080/1828051X.2019.1659106.

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