The effect of fermentation with different times of corn husk which has obtained ammoniation treatment in the production of VFA-NH3 by in vitro digestibility

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Abstract. The objective of the research is to examine the effect of the difference time of fermentation process using *Aspergilus niger* to corn husk that has been ammoniated on fermentability parameter *in vitro* which included the production of VFA and NH_3. The aimed of this research was to determine the proper processing technique for corn husk in an effort to improve its quality as a complete feed ingredient for beef cattle. The experimental design used was completely randomized design with 3 treatment (different time of fermentation : 0; 1 and 2 weeks) and 4 replication. Corn husk which has been ammoniated by 6% ammonia with the temperature of 60 °C for 3 days, added 5% of *Aspergilus niger* as starter, than fermentation with different time. Parameter observed were fermentability parameter **in** *vitro* including the production of VFA and NH_3. Data was analyzed using analysis of variance and if there was significant effect continued by Duncan's test. The results indicate that the longer the fermentation process, the production of VFA and NH_3 increased ($P<0.05$). The conclusion obtained is that the fermentation process using starter of *A. niger* 5% to 2 weeks can increase the production of VFA, NH_3. The best production of VFA and NH_3 occurs in 2 weeks, namely: 122.42 ± 4.35 and 5.99 ± 0.53 mm.

1. Introduction

Effort to increase the availability of feed and an effort for feed cost efficiency can be done by finding new feed sources that have not been used or not commonly used by farmers (unconventional feed ingredients). Some types of agricultural and plantation waste that are not commonly used as food are found in several regions such as corn husk, corncob, soybean peel, cassava peel, cacao peel, peanut skin and coffee peel, but in some other areas has already used these ingredients as animal feed [1].

One of the remaining food crops and plantations that has considerable potential are corn cob and corn husk. The potential of cob as feed has been widely studied, among others, by Tampoebolon [2,3,4], while the corn husk is still not widely studied. The area of harvested corn in Central Java Province in 2015 was 542.804 ha, with corn seed yield of 3,212,391 tons and corn husk waste was approximately 432,387 tons [5]. Basymeleh [6], reported that the corn waste consisted of 50% stems, 20% leaves, 20% cobs, 10% husks. Most of the waste has not been utilized. The corn husk is the outer husk of corn which wraps the corn and usually thrown away. The use of corn husk as a feed has an obstacle because the corn husk has low crude protein content (2.8%) and high crude fibre (36.52%) [7]. Therefore, in its utilization as the feed ingredient, the corn husk needs to be improved its quality, one of them uses ammonia-fermentation processing technology (*Amofer*).

One of the important functions of ammoniation is able to stretch and or to break lignocellulose and hemicellulose bonds while microbial starter can reduce crude fibre level while increasing digestibility and crude protein ingredient. The fermentation process, among others, aims to produce a product (feed ingredient) that has better nutritional and biological availability [8]. The use of ammonia-fermentation technology can increase the crude protein content of corn husk, reduce the crude fibre content and increase the digestibility of corn husk so that it can be used as a good alternative as ruminant animal feed.

Volatile fatty acid (VFA) is the main energy source for the ruminant's body needs, especially beef cattle [9, 10]. The optimum production of VFA from a digestion of feed ingredient is needed to support the productivity of livestock. Tillman *et al*. [11], states that the increased digestibility of feed ingredient will increase VFA production. The feed protein is transformed by bacterial proteolytic enzymes and rumen protozoa to be peptides, amino acid and further transformed to be ammonia (NH_3) [11, 12]. Ammonia production which is high enough from a degradation of feed ingredient will be able to supply the nitrogen needs of rumen microbe which will eventually be utilized by cattle itself [13]. This research aims to examine the effect of the ripening time of fermentation process toward amoniated corn husk on fermentability parameter in vitro including the production of VFA (volatile fatty acid) and NH_3.

2. Materials and methods

The material used were corn husk, *Aspergilus niger* (A. niger), mineral solution for *A. niger* growth according to "American Association of Textile Chemist and Colorist Mineral Salt Iron" (3 g $(NH_4)_2$ NO3, 2.5 g KH_2 PO_4, 2 g K_2 HPO_4, 0.2 g $MgSO_4.7$ H_2O, 0,1 g Fe $SO_4.7$ H_2O), aquades, vinegar (CH_3 COOH) and rice, and one chemical kit for testing *in vitro* parameter including the production of VFA and NH_3. The research equipment used including fermenter, autoclave, thermometer, blender, weight with the capacity of 2 kg with accuracy limit of 10 g, analytical weight with the capacity of 120 g with accuracy of 0.0001 g, universal pH indicator, and one unit of equipment for fermentability testing in vitro includes the production of VFA and NH_3.

The research is divided into 3 phases. The research activity of phase I started with high-temperature ammoniation (60^{o}C) of corn husk using 6% of ammonia level with 3 days of curing period. The research activity of phase II is to carry out the fermentation process using the *Aspergillus niger* as starter with level of 5%. The experimental design used was completely randomized design with 3 treatment (different time of fermentation: 0; 1 and 2 weeks) with 4 replication. The research activity of phase III is to conduct laboratory analysis of fermentability parameter *in vitro* including the production of VFA (volatile fatty acids) and NH_3. The analysis in the test of *in vitro* was carried out according to the Haris method [14].

Data from the observation of each parameter was analyzed using analysis of variance (F Test) and if there was a real effect followed by the DMRT test (Duncan's Multiple range test) according to Steel and Torrie [15].

3. Results and discussion

3.1. Production of Volatyl Fatty Acid (VFA)

The data in Table 1. shows that the time of fermentation have significant effect to VFA production ($P < 0.05$). Volatile Fatty Acid production increased with increasing time of fermentation. The increase in VFA production is mainly due to an increase in organic matter content, mostly in the form of carbohydrate as the main source of VFA. The average VFA production of corn husk the treatment is ranged from 81.67±0.82 to 122.42±4.35 mM, with median value of 101.25 mM.

The production of VFA is sufficient to support the maximum rumen microbial protein synthesis. Optimal production of VFA is to support the maximum rumen microbial protein synthesis, according to Sutardi *et al*. [13] ranges from 80 - 160 mM. The results of the analysis of digestibility of organic material in this research respectively are 37.39% for T_0, 44.80% for T_1 and 60.10% for T_3. The high

digestibility of organic material in fermented corn husk shows that the fermentability of the material is high as well. The more fermentable a feed ingredient, the higher the production of VFA [11]. The optimum production of VFA from a digestion of feed ingredient is really needed to support the livestock productivity.

Table 1. VFA and ammonia production from corn husk fermentation

Parameter *In vitro*	T_0 (0 week)	T_1 (1 week)	T_2 (2 weeks)
VFA (mM)	81.67 ± 0.82^c	99.67 ± 6.22^b	122.42 ± 4.35^a
NH_3 (mM)	3.98 ± 0.07^b	4.11 ± 0.16^b	5.99 ± 0.53^a

[a,b,c] Different superscript in the same row shows a significant difference ($P < 0.05$)

3.2. Production of NH_3

The results of the variance analysis in table 1. show that the treatment have significant effect ($P < 0.05$) to the production of NH_3. The improvement of production of NH_3 *in vitro* along with the increasing time of fermentation. The factors that determine the production of rumen NH_3, among others, are the nitrogen or crude protein content of the fibrous feed ingredient, and its degradability. The protein degradation is also reflected in the digestibility of organic material of the feed ingredient because protein is a component of the organic material. The results of the analysis of crude protein level in the treatment group of T_0, T_1 and T_2 are: 3.94; 5.49 and 8.59%. This increase in NH_3 production is evidenced by the increased digestibility of organic material for longer time of fermentation (2 weeks). The average NH_3 production of corn husk the treatment is ranged from 3.98±0.07 to 5.99±0.53 mM, with an average median value of 4.70 mM. The average production of NH_3 is sufficient to support the rumen microbial protein synthesis. According to Sutardi *et al*. [13], the concentration of optimal N-NH_3 to support the rumen microbial protein synthesis is 3.5 - 7.14 mM.

4. Conclusion

Based on the results of the research, it can be concluded that the treatment of time of fermentation process using *A. niger* 5% to 2 weeks can increase the production of VFA and NH_3.

5. Acknowledgment

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