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# Nutritive value and *in vitro* digestibility of yeast-fermented corn dust with cassava pulp affected by ensiling time

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ABSTRACT: The aims of this study was to determine the chemical composition and nutritive value of yeast-fermented corn dust with cassava pulp using in vitro gas production technique. The experiment followed the completely randomized design with 6×5 factorial arrangement. Factor A comprised 6 ratios of fermented corn dust with cassava pulp (100:0, 80:20, 60:40, 40:60, 20:80, and 0:100% dry matter (DM), respectively), and factor B comprised 5 ensiling times (0, 5, 10, 15, and 20 days ensiled, respectively). Corn dust and cassava pulp were taken to treat with yeast solution according to the treatments and ensiled in a plastic bag at room temperature. The feed samples (200 mg dry matter from each) were incubated in vitro with rumen fluid taken from two male crossbred beef cattle (Thai-native x Charolais) at 2, 4, 6, 8, 12, 18, 24, 36, 48, 72, and 96 h. The results showed that increasing level of cassava pulp as a substrate fermented with corn dust decreased dry matter, crude protein, ether extract, neutral detergent fiber, and acid detergent fiber (P<0.01); whereas, organic matter and crude fiber were increased when increasing the level of cassava pulp (P<0.01). After 15 days ensiled, all treatments of yeast-fermented corn dust with cassava pulp showed greater chemical composition than other ensiling times. Moreover, gas kinetics, gas production, in vitro digestibility, and metabolizable energy content in yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled were significantly greater than other treatments (P<0.01). Therefore, yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM after 15 days ensiled could be an alternative animal feed resource for ruminants.

Keywords: cassava pulp, corn dust; chemical composition; gas production; yeast solution

# Introduction

Corn dust and cassava pulp are by-products remaining after corn milling and cassava starch processing which has potential as an alternative feed resource for ruminant production. The utilization of corn dust and cassava pulp would be beneficial since the material is abundantly and available in many animal feed factories and cassava processing factories. In Thailand, 4.5-5.0 million tonnes of corn are produced during 2018-2020 (Office of agricultural economics, 2021), and approximately 2% of corn dust is produced during the corn milling process. Corn dust comprises cracked

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corn kernels, dust, cob and hulls, which contains 8.4-8.8% crude protein (CP), 40.7-42.6% neutral detergent fiber (NDF) and 16.4-17.0% acid detergent fiber (ADF), on dry matter (DM) (Lunsin et al., 2021). Whereas, 1.5–2.0 million tonnes of cassava pulp are produced annually from the entire cassava starch industry (Chauynarong et al., 2015). Fresh cassava pulp contains 15.8–23.4% DM with 1.2–2.8% CP, 55.0–74.4% nitrogen-free extract (NFE), and 17.9–24.0% crude fiber (CF), on DM basis (Yimmongkol, 2009). Recently, the price of corn dust and cassava pulp is always cheap approximately 3.0 baht/kg of corn dust and 0.27 baht/kg of fresh cassava pulp. Therefore, corn dust and cassava pulp are a potential feed resource to reduce the production cost of livestock. They represent valuable biomass and potential supply of protein and energy if appropriate technologies can be deployed for nutrient enrichment.

Fermentation is one of the major technologies for nutrient enrichment of agro-wastes. It is the best conserving technique because it is less expensive and easy to make, which also has the advantage of minimum loss of nutrients and increase of the quality of feed stuff (Hao et al., 2021; Ubalua, 2007, Sadh et al., 2018). Several studies had been successful in improving the nutritional value of cassava pulp and used as animal feed. A previous study found that yeast (Saccharomyces cerevisiae) fermented cassava pulp could increase protein content of cassava pulp (Khampa et al. 2009) and improved ruminal undegradable protein (RUP) content (Kaewwongsa et al., 2011). Cassava pulp could be fermented with yeast (S. cerevisiae) or a mixture of microbes (effective microorganisms, EM) in combination with molasses and urea (MU), in order to be conserved and used as animal feed. Thus, the nutritive value of fermented cassava pulp with yeast and EM was improved by MU supplementation (Pilajun and Wanapat, 2018). In addition, urea and molasses treatment could improve the nutritive value of treated cassava pulp, which increase CP and decrease fiber contents (Norrapoke et al., 2018). As for corn dust, Lunsin et al. (2021) reported that the nutritive value of corn dust could be enhanced by urea and molasses treatment. Urea treatment ensured the quality of ensiled corn dust by increasing CP and reducing fiber content, whereas molasses treatment improved organic matter (OM) content of ensiled corn dust. Adding 4% urea in combination with 2-6 % molasses are the best options for improving the nutritive values of corn dust as silage for feeding ruminants. Based on the literature review, the fermentation of corn dust and cassava pulp not only changes nutrient composition but also results in preservation of the fermented products, reduced time of degradation and improve feed utilization.

Although corn dust has a relatively high content of fiber (NDF and ADF), it remains a source of protein and energy. Whereas, fresh cassava pulp consists of 50% starch (Chauynarong et al., 2015), small particles and less physically effective NDF (Chumpawadee and Leetongdee, 2020) which is a source of non-fibrous carbohydrate. Thus, yeast-fermented corn dust with cassava pulp is an alternative technology to increase the nutritive value of corn dust and cassava pulp which has the potential supply of protein and energy and improve productivity of ruminants as well as to control feed cost. Additionally, there is very little information available on the corn dust chemical composition and silage fermentation profile, and the reports of corn dust conserving, and using as ruminant feed are also lacking. It could be hypothesized that the yeast-fermented corn dust in combination with cassava pulp could potentially improve the nutritive value and *in vitro* digestibility of fermented corn dust with cassava pulp, which was potentially supplying protein and energy feed resources for ruminants. Therefore, this study aimed to determine the effects of yeast-

fermented corn dust with cassava pulp on chemical composition and *in vitro* fermentation using gas production techniques.

#### Materials and methods

This experiment was conducted under Feed Analysis Laboratory, and Animal Biotechnology and Reproduction Laboratory, Faculty of Agriculture, Ubon Ratchathani Rajabhat University, Thailand. Experimental protocols and animal care were approved by the ethics committee of Ubon Ratchathani Rajabhat University (Approval no. AN61007) according to the guidelines of Ethical Principles for the Use of Animals for Scientific Purposes of the National Research Council of Thailand (NRCT).

# Experimental design and treatments preparation

The experiment followed the Completely Randomized Design (CRD) with 6x5 factorial arrangement of treatments and three replicates were performed for all treatments. Factor A comprised 6 ratios of fermented corn dust with cassava pulp (100:0, 80:20, 60:40, 40:60, 20:80, and 0:100% DM, respectively), and factor B comprised 5 ensiling times (0, 5, 10, 15, and 20 days, respectively).

Corn dust was collected from Kao Na Animal Food Industry Company Limited (Kao Na Animal Food Industry Co., Ltd., Thailand) located in Samrong District, Ubon Ratchathani Province. Fresh cassava pulp was collected from Eimsiri Starch Company Limited (Eimsiri Starch Co., Ltd., Thailand) Kantharalak District, Si Sa Ket Province. Corn dust and fresh cassava pulp was taken to treated with yeast solution based on the method described by Boonnop et al. (2009) and Khampa et al. (2011). In brief, the methods of yeast solution were modified by dissolved 200 g of yeast (Saccharomyces cerevisiae) with 200 g of sugar in 1000 ml of distilled water, then mixed and incubated at room temperature for 10 min. (A). Medium solution was prepared by dissolved 600 g of molasses and 200 g of urea in 10 l of distilled water, and incubated at room temperature for 10 min. (B). Then, remove yeast media solution (A) into a medium (B) and continue incubated at room temperature for 24h. After 24h of incubation, then transfer 10 l of yeast media solution mixed with 10 kg (DM basis) of the treatment of fermented corn dust with cassava pulp at the ratio of 100:0, 80:20, 60:40, 40:60, 20:80, and 0:100% DM, respectively. The mixtures were carefully packed into the plastic bags and compressed by hand to remove as much air as possible. Each bag was tightly tied and put into a second empty plastic bag which was also tied to protect it from rupturing and left undisturbed at room temperature until the period of sampling collection in different ensiling times (0, 5, 10, 15, and 20 days, respectively).

### Sample collection and analysis

Samples of corn dust, cassava pulp and all treatment of yeast-fermented corn dust with cassava pulp were collected after 0, 5, 10, 15, and 20 days ensiled, respectively. Collected samples were dried by hot air oven at 60 °C for 48 h then ground to pass a 1 mm sieve for chemical analysis and *in vitro* gas production procedure. Chemical composition of dry matter (DM), ash, crude protein (CP) and ether extract (EE) were analysed by the proximate analysis

procedure according to AOAC (1990). Fiber contents including neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined using the detergent analysis method of Van Soest et al. (1991).

# In vitro gas production technique

Two male, crossbred Thai-native x Charolais beef cattle (350±20 kg body weight) were used as rumen fluid donor. The animals were fed individually penned, clean fresh water and mineral blocks were offered as free choice. The animals were fed with rice straw as a roughage on ad libitum basis and concentrate (14% crude protein, 2.5 Mcal/ kg of ME) was fed at 0.5% body weight in two equal portions, at 0700 am and at 0400 pm. The animals were given the diets for 14 days before the rumen fluid was collected. The 1000 ml rumen fluid was obtained from each animal by using a stomach tube connected with a vacuum pump, before morning feeding. The rumen fluid was filtered through four layers of cheesecloth into pre-warmed thermo flasks. A strict anaerobic technique was used and during of the rumen fluid collection according to the method of Menke et al. (1979). Preparation of artificial saliva was done according to Menke and Steingass (1988). The artificial saliva and rumen fluid were mixed at a 2:1 ratio to produce a rumen inoculation mixture. Three bottles containing only rumen inoculation mixture were used as blank. Mean gas production of the blank samples was subtracted from each measurement to give the net gas production. The glass bottles with 200 mg of substrate treatments after 0, 5, 10, 15 and 20 days ensiled, were pre-warmed in a water bath at 39°C before filling with 30 ml of rumen inoculation mixture. Bottles were sealed with rubber stoppers and aluminum caps and incubated at 39°C for in vitro gas test. The volume of gas production was recorded on 2, 4, 6, 8, 12, 18, 24, 36, 48, 72, and 96h of incubation. Cumulative gas production data was fitted to the model of Ørskov and McDonald (1979) as follow:

$$y = a + b (1 - e^{-ct})$$

Where: y = gas produced at time 't'; a = the gas production from the immediately soluble fraction (ml); b = the gas production from the insoluble fraction (ml); c = the gas production rate constant for the insoluble fraction (b) (%/h); t = the gas production time; and lal+b is the potential extent of gas production (ml).

At 24 h post inoculation a set of three replicate samples were taken to determine *in vitro* degradability, when the contents were filtered through pre-weighed Gooch crucibles and residual dry matter was estimated. The percent loss in weight was determined and presented as *in vitro* dry matter degradability (IVDMD). The dried feed sample and residue left above were ash at 550 °C for determination of *in vitro* organic matter degradability (IVOMD) (Tilley and Terry, 1963). Calculations were made using the following equation:

IVDMD (%)= 
$$\frac{\text{(WD -WC)} - \text{(WB-WC)}}{\text{W/S}} \times 100$$

Where WD = weight of the crucible and the residue after drying at 100°C, WB = weight of the crucible and the chemical reagent residue after drying at 100°C (blank), WC = dry weight of crucible, and WS = dry weight of original sample (DM basis).

The volume of gas produced after 24h of incubation was used with CP, EE, and ash content to estimate metabolizable energy (ME, MJ/kg DM) based on the following equation for concentrate as reported by Menke and Steingass (1988):

ME 
$$(MJ/kg DM) = 1.06 + (0.157GP) + (0.0084CP) + (0.022EE - 0.0081ash)$$

Where: ME = metabolizable energy (MJ/kg DM), GP = gas production at 24h incubation (ml/200 mg DM), CP=crude protein (%), and EE = ether extract (%).

# Statistical analysis

All data were subjected to analysis of variance (ANOVA) according a 6×5 factorial arrangement in a CRD using the General Linear Model of SAS (2006). Data were analyzed using the following model:

$$Y_{iik} = \mathcal{L} + A_i + B_i + AB_{ii} + \mathcal{E}_{iik}$$

Where  $Y_{ijk}$  = observations;  $\mu$  = overall mean;  $A_i$  = effect of factor A (Corn dust to cassava pulp ratio, i = 100:0, 80:20, 60:40, 40:60, 20:80, and 0:100% DM, respectively);  $B_j$  = effect of factor B (ensiling times, j = 0, 5, 10, 15 and 20 days, respectively),  $AB_{ij}$  = interaction between factor A and B, and  $E_{ijk}$  = the residual effect. Orthogonal contrast was used to investigate the effect of the treatments respond. Differences among mean was tested by Duncan's New Multiple Range Test (DMRT) (Steel and Torrie, 1980) with P<0.05 was accepted as representing statistically significant differences.

#### Results and Discussion

## Chemical composition

Corn dust contained 94.4, 91.0, 8.4, 12.9, 2.8, 40.7 and 16.4% of DM, OM, CP, CF, EE, NDF, and ADF, respectively. Whereas, fresh cassava pulp contained 23.1, 92.4, 1.8, 19.1, 0.2, 29.6 and 14.3% of DM, OM, CP, CF, EE, NDF, and ADF, respectively (**Table 1**). It was found that the CP content of corn dust in this study was similar to Lunsin et al. (2021) who found that the CP content in corn dust was 8.4-8.8% DM, whereas cassava pulp with a CP content of 2.2-2.5% DM was reported by Keaokliang et al. (2018) which was comparable to 1.8% CP found in this study. This may be due to the variety of cassava, rate of fertilizer application and other crop management factors, as well as starch extraction methodology of each factory (Pilajun and Wanapat, 2018)

Table 1 The nutritive value of corn dust and cassava pulp

ltem	DM	ОМ	СР	CF	EE	NDF	ADF
	(%)	(% DM)					
Corn dust	94.4	91.0	8.4	12.9	2.8	40.7	16.4
Cassava pulp	23.1	92.4	1.8	19.1	0.2	29.6	14.3

DM = dry matter, OM = organic matter, CP = crude protein, CF = crude fiber, EE = ether extract, NDF = neutral detergent fiber, ADF = acid detergent fiber.

The chemical composition of yeast-fermented corn dust with cassava pulp are presented in Table 2. The interaction between the ratio of corn dust to cassava pulp and ensiling time on chemical composition was found to be statistically significant (P<0.01). The increasing levels of cassava pulp as a substrate in yeast-fermented corn dust with cassava pulp resulted in lower DM, CP, EE, NDF, and ADF contents because cassava pulp has low contents of DM, CP, EE, NDF, and ADF. The lowest chemical composition of experimental diet was found in the treatment of yeast-fermented 0% DM of corn dust with 100% DM of cassava pulp or yeast-fermented cassava pulp. In contrast, the OM and CF contents were increased due to the different levels of yeast-fermented corn dust with cassava pulp, and OM recorded were 89.6, 89.9, 91.1, 91.8, 91.3, and 92.0% DM, respectively; while CF contents of 12.1, 13.3, 14.7, 15.9, 16.9, and 18.4% DM were observed. Moreover, at day 0 of ensiled (unfermented treatments) the chemical composition of experimental diets was lower than that of day 5, 10, 15, and 20 of ensiled (P<0.01). The CP content of yeast-fermented corn dust with cassava pulp was increased due to the different ensiling time, especially on days 15 and 20 of ensiled, the highest CP recorded were 12.7 and 12.5% DM, respectively. Whereas, increasing ensiling time influenced on decrease fiber fractions (CF, NDF, and ADF contents). The effected of ensiling time on chemical composition in the current study agreed with Lunsin et al. (2018; 2019) who indicated that after the sugarcane bagasse had been fermented with urea and molasses substrate at 21d of fermentation period, the nutritive value and in vitro fermentation could be improved. According to Sadh et al. (2018) noted that fermentations used for biotransformation of agro-wastes into valuable products having low cost and high nutritive value. Additionally, the nutrient contents of experimental diet were increased as affected by solid state fermentation with yeast. Ajila et al. (2012) reported that solid state fermentation (fungi, yeast and bacteria) of agro-industrial residues has been proposed as a suitable pretreatment for protein enrichment that can lead its use as animal feed. The effect could be attributed to the possible secretion of some extracellular enzymes into the fermented products by fermenting microorganisms and to the addition of urea. Apart from this, the increase in the growth and proliferation of the fungi/yeast complex in the form of single cell proteins may possibly account for the apparent increase in the CP content of the products (Suksombat et al., 2018). As shown in Table 1 and 2, the CP content of corn dust (unfermented) was increased from 8.4% DM to 13.5% DM by fermented corn dust with yeast solution (yeast-fermented 100% DM of corn dust with 0% DM of cassava pulp or yeast-fermented corn dust). Also, the CP content of cassava pulp (unfermented) was increased from 1.8% DM to 8.5% DM in the treatment of yeast-fermented cassava pulp (yeast solution fermented 0% DM of corn dust with 100% DM of cassava pulp). An improved nutrient composition of agro-industrial by-products caused by solid state fermentation was also noted in other studies. For example, the co-culture of Candida utilis and Aspergillus niger increased the protein content of dried and pectin-extracted apple pomace to 20% and 17%, respectively, under solid state fermentation conditions (Bhalla and Joshi, 1994). The protein content of cassava pulp was increased by fermentation with S. cerevisiae (Khampa et al., 2011). Fermentation of cassava products using Aspergillus oryzae increased the amounts of reducing sugar and fermentation of cassava products using A. oryzae and S. cerevisiae increased CP content (Suksombat et al., 2018). Also, Akindahunsi and Oboh (2003) indicated that fungi fermentation of cassava mash significantly increased (P<0.05) the in vitro multienzyme protein digestibility of the cassava products.

Table 2 Chemical composition of yest-fermented corn dust with cassava pulp as affected by ensiling time (% DM)

Treatment	DM	OM	CP	CF	EE	NDF	ADF
	(%)			(% [	DM)		
Corn dust : cassava pulp, % DM (A	4)						
100:0	52.6 <sup>a</sup>	89.6 <sup>c</sup>	13.5 <sup>a</sup>	12.2 <sup>f</sup>	3.0 <sup>a</sup>	34.6°	14.1ª
80 : 20	40.0 <sup>b</sup>	89.9 <sup>c</sup>	13.0 <sup>b</sup>	13.3 <sup>e</sup>	2.7 <sup>b</sup>	32.5 <sup>b</sup>	13.8 <sup>b</sup>
60 : 40	32.7 <sup>c</sup>	91.1 <sup>b</sup>	12.6 <sup>c</sup>	14.7 <sup>d</sup>	2.0°	31.6°	13.6 <sup>bc</sup>
40 : 60	27.3 <sup>d</sup>	91.8°	11.8 <sup>d</sup>	15.9 <sup>c</sup>	1.5 <sup>d</sup>	30.9 <sup>d</sup>	13.5°
20 : 80	24.9 <sup>e</sup>	91.3 <sup>b</sup>	10.4 <sup>e</sup>	16.9 <sup>b</sup>	0.8 <sup>e</sup>	29.8 <sup>e</sup>	13.3 <sup>d</sup>
0:100	21.9 <sup>f</sup>	92.0 <sup>a</sup>	8.5 <sup>f</sup>	18.4ª	0.3 <sup>f</sup>	26.2 <sup>f</sup>	12.9 <sup>e</sup>
Ensiling time, days (B)							
0	36.2°	90.2 <sup>c</sup>	10.0 <sup>d</sup>	16.0°	1.3 <sup>c</sup>	31.8 <sup>a</sup>	14.9ª
5	33.2 <sup>b</sup>	90.5 <sup>c</sup>	10.7 <sup>c</sup>	15.4 <sup>b</sup>	1.8 <sup>b</sup>	30.9 <sup>b</sup>	14.2 <sup>b</sup>
10	31.9 <sup>c</sup>	90.9 <sup>b</sup>	12.2 <sup>b</sup>	15.3 <sup>b</sup>	1.8 <sup>b</sup>	30.5 <sup>c</sup>	13.0°
15	32.1 <sup>bc</sup>	91.7ª	12.7ª	14.7°	1.9 <sup>a</sup>	30.4 <sup>c</sup>	12.9 <sup>c</sup>
20	32.9 <sup>bc</sup>	91.3 <sup>b</sup>	12.5 <sup>a</sup>	14.6°	1.9 <sup>a</sup>	30.3 <sup>c</sup>	12.8 <sup>c</sup>
SEM	0.57	0.52	0.57	0.53	0.57	0.57	0.55
Comparison							
A	**	**	**	**	**	**	**
В	**	**	**	**	**	**	**
AxB	**	**	**	**	**	**	**
Orthogonal polynomial							
A (lin)	**	**	**	**	**	**	**
A (quad)	**	**	**	ns	*	**	ns
A (cubic)	**	ns	**	ns	**	**	ns
A (quar)	ns	**	*	**	ns	**	ns
B (lin)	**	**	**	**	**	**	**
B (quad)	**	ns	**	*	**	**	**
B (cubic)	ns	**	**	ns	**	ns	**
B (quar)	ns	ns	**	**	**	ns	**

DM = dry matter, OM = organic matter, CP = crude protein, CF = crude fiber, EE = ether extract, NDF = neutral detergent fiber, ADF = acid detergent fiber, SEM = standard error of the mean, Orthogonal polynomial; lin = linear, qua = quadratic, cub = cubic, qua = quartic, a,b,c,d,e,f = Mean within columns with different superscript letters differed (P<0.05), \*P<0.05, \*\*P<0.01, ns = non-significant.

#### *In vitro* rumen fermentation

The gas production characteristics of corn dust and cassava pulp are presented in Table 3. It was found that the gas kinetics in term of gas production from the immediately soluble fraction (a), gas production from the insoluble fraction (b), the rate of gas production (c), potential extent of gas production (lal+b) and cumulative gas production (ml/200 mg DM) at 24, 48, 72, and 96h of incubation times, and in vitro digestibility of DM and OM (IVDMD and IVOMD), also metabolizable energy (ME) in cassava pulp were higher than that of corn dust. In this study, the negative a intercept value of -4.9 and -9.5 ml were observed in corn dust and cassava pulp. The negative a intercept found in this study indicated a lag phase due to delay in microbial colonization of the substrate may occur in the early stage of incubation (Chumpawadee et al., 2005; 2006). The gas production from the insoluble fraction (b) of corn dust and cassava pulp were 58.5 and 70.8 ml, respectively. The high fermentation of the insoluble fraction was observed in cassava pulp, possibly influenced by the carbohydrate fractions readily available to the microbial population (Chumpawadee et al., 2007). Moreover, high rate of gas production (c, %/h) and potential extent of gas production (lal+b) were observed in cassava pulp (0.068 %/h and 80.3 ml, respectively), indicated high rate of degradation and fermentation in the rumen of cassava pulp. In addition, the IVDMD and IVOMD of corn dust were 49.6 and 63.8%, respectively, and cassava pulp were 65.9 and 71.4%, respectively. The calculated amounts of ME of corn dust and cassava pulp were 16.7 and 20.8 MJ/kg DM, respectively. The different chemical composition of corn dust and cassava pulp had distinct gas production and digestibility. Corn dust had more fiber, and higher CP content than cassava pulp; these different chemical composition influence on lower digestibility performance. In contrast, cassava pulp had higher digestibility than corn dust, mainly attribute to its lower fiber content (Getachew et al., 2004).

**Table 3** Kinetic of gas production, gas production volume (ml/200 mg DM), *in vitro* degradability (%) and metabolizable energy (MJ/kg DM) of corn dust and cassava pulp

		Gas kinetics			C25.1/6	Cas valuma (ml/200 mg DM)				In vitro		
Item			Gas VC	Gas volume (ml/200 mg DM)				degradability (%)				
	а	b	С	lal+b	24h	48h	72h	96h	IVDMD	IVOMD	(MJ/kg DM)	
	(ml)	(ml)	(%h)	(ml)								
Corn d	ust											
	-4.0	58.5	0.036	62.5	41.5	50.1	52.3	53.4	49.6	63.8	16.7	
Cassav	a pulp											
	-9.5	70.8	0.068	80.3	77.5	78.7	78.9	79.0	65.9	71.4	20.8	

a = the gas production from the immediately soluble fraction (ml), b = the fermentation of the insoluble fraction (ml), c = rate of gas production (%/h), lal+b = potential extent of gas production (ml), IVDMD =  $in\ vitro$  dry matter degradability (%), IVOMD =  $in\ vitro$  organic matter degradability (%), ME = Metabolizable energy (MJ/kg DM).

Kinetics of in vitro gas production, cumulative gas production (ml/200 mg DM) in different incubation times, in vitro digestibility, and ME (MJ/kg DM) of yeast-fermented corn dust with cassava pulp are presented in Table 4 and 5. Gas production volumes, gas kinetic parameters, in vitro digestibility, and ME of different ratios of corn dust to cassava pulp were differed significantly (P<0.01) among treatments as affected by ensiling time. Increasing the level of cassava pulp as a substrate in yeast-fermented corn dust with cassava pulp could increase the gas production volumes and gas kinetic parameters (a, b, c, and lal+b), which was highest when using cassava pulp 60-100% DM as a substrate fermented with corn dust after 15 days of ensiled (Table 4). In terms of the gas production kinetics, a is the intercept and ideally reflects the fermentation of the soluble and readily available fraction of the feed, b describes the fermentation of the insoluble (but with time fermentable) fraction and c the fractional rate at which b is fermented per h, and a+b describes total fermentation (Blümmel and Becker, 1997). The highest values for these parameters were observed in yeastfermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled, indicated the highest fermentation of soluble fraction and the insoluble fraction of the feeds, as well as the fastest rate of gas production, possibly influenced by the soluble carbohydrate fraction readily available to the rumen microbes (Chumpawadee et al., 2007; Akinfemi et al., 2009). Moreover, yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled were determined to have a higher potential extent of gas production (lal+b, ml) values than other treatments. These could be due to increased cassava pulp levels that would provide more readily available energy, enhancing the corresponding of microbes, consequently, increased total digestion and fermentation of the feeds.

Likewise, the gas production volumes at different incubation times and *in vitro* digestibility of yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled was higher than those other ensiling times (P<0.01) (Table 5). Different gas production in these studies can be due to different chemical constituents of experimental diet. There was a positive correlation between non-fiber carbohydrate (NFC) content of feeds and gas production, but CP and NDF levels were negatively correlated with gas production (Menke and Steingass, 1988; Getachew et al., 2004). Also, there are significant correlation between *in vitro* gas measurement and *in vivo* digestibility. Menke and Steingass (1988) noted that there was a positive correlation between ME calculated from *in vitro* gas production together with CP and fat content with ME value of conventional feeds measured *in vivo*. Additionally, the relatively high content of cell wall structures (hemicelluloses and cellulose) might restrict microorganism activity, and then lowered gas volume (Idris et al., 2011). In consistent with our study, increasing level of cassava pulp as a substrate fermented with corn dust could decrease CP and NDF content in the diet, resulted in an increase in gas production, *in vitro* digestibility and ME content. Increasing level of cassava pulp in yeast-fermented corn dust with cassava pulp revealed an increase in gas production at 24h incubation, which was highest in the treatment of yeast-fermented 0% DM of corn dust with 100% DM of cassava pulp (yeast-fermented cassava pulp), so it recorded the highest energy content.

Table 4 Kinetic of gas production of yeast-fermented corn dust with cassava pulp as affected by ensiling time

Treatment	Gas kinetics						
	a (ml)	b (ml)	c (%h)	lal+b (ml)			
Corn dust : Cassava pulp, % DM (A)							
100 : 0	-4.0 <sup>a</sup>	61.3 <sup>d</sup>	0.046 <sup>e</sup>	65.3 <sup>d</sup>			
80 : 20	-4.7 <sup>b</sup>	62.8 <sup>c</sup>	0.049 <sup>d</sup>	67.4°			
60 : 40	-6.5 <sup>c</sup>	69.4 <sup>b</sup>	0.051 <sup>d</sup>	75.8 <sup>b</sup>			
40 : 60	-7.3 <sup>d</sup>	71.4ª	0.053 <sup>b</sup>	78.6ª			
20:80	-7.4 <sup>d</sup>	71.6ª	0.055 <sup>a</sup>	79.0°			
0:100	-7.6 <sup>d</sup>	72.2 <sup>a</sup>	0.055 <sup>a</sup>	79.8 <sup>a</sup>			
Ensiling time, days (A)							
0	-6.0 <sup>a</sup>	63.7 <sup>d</sup>	0.049 <sup>c</sup>	69.8 <sup>d</sup>			
5	-6.2 <sup>ab</sup>	64.5 <sup>d</sup>	0.052 <sup>b</sup>	70.6 <sup>d</sup>			
10	-6.4 <sup>bc</sup>	68.1°	0.052 <sup>b</sup>	74.4 <sup>c</sup>			
15	-6.5 <sup>b</sup>	73.0°	0.053 <sup>a</sup>	79.5°			
20	-6.0 <sup>a</sup>	71.2 <sup>b</sup>	0.051 <sup>b</sup>	77.3 <sup>b</sup>			
SEM	0.54	0.54	0.53	0.54			
Comparison							
A	**	**	**	**			
В	ns	**	**	**			
$A \times B$	**	ns	**	ns			
Orthogonal polynomial							
A (lin)	**	**	**	**			
A (quad)	**	**	**	**			
A (cubic)	*	*	ns	*			
A (quar)	××	**	ns	**			
B (lin)	ns	**	*	**			
B (quad)	**	*	**	**			
B (cubic)	*	**	ns	**			
B (quar)	ns	ns	**	ns			

a = the gas production from the immediately soluble fraction (ml), b = the fermentation of the insoluble fraction (ml), c = rate of gas production (%/h), lal+b = potential extent of gas production (ml), IVDMD = *in vitro* dry matter degradability (%), IVOMD = *in vitro* organic matter degradability (%), SEM = standard error of the mean, Orthogonal polynomial; lin = linear, qua = quadratic, cub = cubic, qua = quartic, a,b,c,d,e = Mean within columns with different superscript letters differed (P<0.05), \*P<0.05, \*\*P<0.01, ns = non-significant.

**Table 5** Gas production volumes (ml/200 mg DM), *in vitro* degradability (%) and metabolizable energy (MJ/kg DM) of yeast-fermented corn dust with cassava pulp as affected by ensiling time

Treatment	Gas	Gas volume (ml/200 mg DM)				egradability %)	Metabolizable energy
	24h	48h	72h	96h	IVDMD	IVOMD	(ME, MJ/kg DM)
Corn dust : Cassa	ava pulp %	DM (A)					
100:0	53.2 <sup>e</sup>	58.7 <sup>e</sup>	59.7 <sup>d</sup>	60.4 <sup>d</sup>	53.0 <sup>d</sup>	66.8 <sup>d</sup>	20.0 <sup>c</sup>
80 : 20	57.8 <sup>d</sup>	62.2 <sup>d</sup>	63.0°	63.5 <sup>c</sup>	55.7 <sup>c</sup>	68.7 <sup>c</sup>	20.4 <sup>bc</sup>
60 : 40	67.3 <sup>c</sup>	71.4°	72.2 <sup>b</sup>	72.6 <sup>b</sup>	60.4 <sup>b</sup>	70.2 <sup>b</sup>	20.7 <sup>b</sup>
40 : 60	71.9 <sup>b</sup>	75.0 <sup>b</sup>	75.6 <sup>a</sup>	76.0 <sup>a</sup>	67.1 <sup>a</sup>	71.0 <sup>ab</sup>	20.6 <sup>b</sup>
20 : 80	72.7 <sup>ab</sup>	75.8 <sup>ab</sup>	76.3 <sup>a</sup>	76.5 <sup>a</sup>	66.8 <sup>a</sup>	70.7 <sup>ab</sup>	21.3ª
0:100	73.5 <sup>a</sup>	76.8 <sup>a</sup>	77.2 <sup>a</sup>	77.4 <sup>a</sup>	67.6 <sup>a</sup>	71.5 <sup>a</sup>	20.7 <sup>b</sup>
Ensiling time, day	ys (B)						
0	59.6 <sup>e</sup>	64.2 <sup>e</sup>	65.0 <sup>e</sup>	65.5 <sup>e</sup>	59.1 <sup>c</sup>	67.4 <sup>d</sup>	20.3 <sup>b</sup>
5	62.3 <sup>d</sup>	66.3 <sup>d</sup>	66.8 <sup>d</sup>	67.1 <sup>d</sup>	60.8 <sup>b</sup>	69.3 <sup>c</sup>	20.4 <sup>ab</sup>
10	66.2 <sup>c</sup>	70.2 <sup>c</sup>	70.9 <sup>c</sup>	71.3 <sup>c</sup>	61.5 <sup>b</sup>	70.2 <sup>b</sup>	20.7 <sup>ab</sup>
15	72.5ª	76.0 <sup>a</sup>	76.7 <sup>a</sup>	77.0 <sup>a</sup>	63.8 <sup>a</sup>	71.5°	20.8 <sup>a</sup>
20	69.2 <sup>b</sup>	73.1 <sup>b</sup>	74.0 <sup>b</sup>	74.4 <sup>b</sup>	63.6 <sup>a</sup>	70.8 <sup>ab</sup>	20.8 <sup>a</sup>
SEM	0.56	0.55	0.55	0.55	0.56	0.51	0.55
Comparison							
Α	**	**	**	**	**	**	ns
В	**	**	**	**	**	**	**
AxB	ns	ns	*	*	**	**	**
Orthogonal polyr	nomial						
A (lin)	**	**	**	**	**	**	**
A (quad)	**	**	**	**	**	**	*
A (cubic)	*	*	*	**	**	ns	ns
A (quar)	**	**	**	*	**	ns	ns
B (lin)	**	**	**	**	**	**	**
B (quad)	**	**	**	**	ns	**	ns
B (cubic)	**	**	**	*	ns	ns	ns
B (quar)	**	*	*	**	*	ns	ns

IVDMD =  $in\ vitro$  dry matter degradability (%), IVOMD =  $in\ vitro$  organic matter degradability (%), SEM = standard error of the mean, Orthogonal polynomial; lin = linear, qua = quadratic, cub = cubic, qua = quartic, a,b,c,d,e = Mean within columns with different superscript letters differed (P<0.05), \*P<0.05, \*\*P<0.01, ns = non-significant.

#### Conclusions

The results of this study demonstrate that increasing levels of cassava pulp as a substrate fermented with corn dust could decrease CP and fiber (NDF and ADF) contents, whereas gas production characteristics, *in vitro* digestibility, and metabolizable energy contents increased. The chemical composition of experimental diets could improve after days 15 of ensiled, the highest gas production characteristics, *in vitro* digestibility and metabolizable energy contents were observed in yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled. Based on this study, it could be concluded that yeast-fermented corn dust with cassava pulp at the ratio of 40:60, 20:80, and 0:100% DM on day 15 of ensiled could improve the nutritive value and *in vitro* rumen fermentation characteristics. Therefore, yeast-fermentation corn dust with cassava pulp is a good feeding technology for utilizing industry by-products and provides a promising feedstuff. Further *in vivo* experiment in feeding trials is needed to be more informative about yeast-fermented corn dust with cassava pulp on the performance of livestock.

#### Conflict of interest

The authors declare that there are no conflicts of interest.

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