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# Effect of Yeast Fermented Ethanol Waste on Feed Utilization and Digestion in Dairy Cattle

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**Abstract:** The industrial production of ethanol from cassava produces a large amount of waste. The residual nutrients in this by-product are enough to supplement ruminant animal feeding. Therefore, the objective of this study was to study the effect of yeast fermented ethanol waste (FEW) on feed efficiency and dairy cow performance. Twelve lactating Holstein Friesian cows were assigned to four levels of FEW 0, 25, 35 and 45% of DM in total mixed ration (TMR) diet with fresh ruzi grass (*Brachiaria ruziziensis*) as the roughage source. Dry matter intake (DMI) and body weight had increased when fed up to 25% of FEW (14.2, 15.4, 11.4 and 7.8 kg/d and 23.7, 6.2, -17.8 and -40.6 kg, respectively; $p<0.01$). While digestibility of DM, CP, tended to increase, there were no significant difference ($p>0.05$) in fat, NDF and ADF digestibility. Milk production was higher in animals fed 0 and 25% FEW of TMR diets compared to 35 and 45% (10.8, 11.1, 9.9 and 8.1 kg/d, respectively; $p<0.05$), while milk composition was not affected ($p>0.05$). There were no effects on ruminal pH and concentrations of ruminal $NH_3$-N, acetate ($C_2$), propionate ($C_3$), butyrate ($C_4$) and the $C_2$:$C_3$ ratio ($p>0.05$). Neither were blood glucose and blood urea nitrogen affected ($P>0.05$). However, total volatile fatty acids (VFA) were reduced with increasing levels of FEW (110.9, 100.9, 105.8 and 94.0 mM/l, respectively; $p<0.05$). Therefore, it can be concluded that FEW up to 25% of DM can be used in TMR diets for dairy cow feeding.

**Key words:** Ethanol waste, fermented, improved feed, yeast, utilization

## INTRODUCTION

Many field crops such as corn, sugar cane and cassava can be used for ethanol production. In Asian countries cassava root is normally used as a main raw material for ethanol production. The cassava root material and ethanol production ratio is 2.3-2.5:1 wt/wt and thus producing the waste of fermented cassava 8-10% of raw material. The ethanol waste from an ethanol plant is estimated to be at least 30 ton for 150 ton ethanol production per day. Ethanol production has been increasing yearly because of global energy demand with proportional increase in ethanol waste which could pose associated problems of waste disposal. To alleviate the problem, there is a need for research to make use of the ethanol waste and one of the potential avenues is its utilization as animal feed. Ethanol waste was found to contain 16.8% DM, 6.1% CP, 48.6% NDF, 34.2% ADF, 1.0% ether extract and 7.8% ash with a pH of 4.2 (KKU-Lab, 2012). Given its low protein and high moisture content with medium energy and a good non-forage fiber, it could be used as an ingredient in ruminant feed. Commercial yeast fermentation of ethanol waste is known to improve protein and digestibility of the waste and is well documented in both *in-vitro* and *in-vivo* studies. Yeast (Saccharomyces cerevisiae) can be supplemented in ruminant diets to favorably modify the ruminal environment and promote microbial growth (Desnoyers *et al*., 2009; Robinson and Erasmus, 2009). Oboh and Akindahunsi (2003) studied the results of fermented cassava pulp with yeast (Saccharomyces cerevisiae) for 3 days and found that the protein content was improved from 4.4 to 10.9% while reducing the cyanide content from 21.3 to 9.5 mg/kg. Lyayi and Losel (2001) fermented cassava pulp with Aspergillus niger for 20 days and found that protein content was increased from 3.6 to 9.0% compared to fermenting with yeast (7.2%). Sunato *et al*. (2012) fermented ethanol waste with yeast, 2% urea and 6% sugar(w/w) for 15 days and the protein content increased to 20 to 25%. Boonnop (2008) found that using yeast fermented cassava chips with 6% urea and 3% molasses (30.4% CP) increased total VFA from 104.8 to 116.3 mM/L and total bacteria increased from $2.6 \times 10^{10}$ to $6.4 \times 10^{10}$ cell/ml and concluded that it could replace soybean meal in a concentrate diet fed at 1% of body weight in male dairy cattle. Consequently, the objective of this study was to determine the optimum levels of yeast fermented ethanol waste (FEW) on feed intake, rumen fermentation and performance of dairy cattle.

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## MATERIALS AND METHODS

**Animals and treatments:** Twelve 98.4% purebred Holstein Friesian lactating dairy cows with average body weight of 405.4±43.9 kg) in mid-lactation (average milk yield 10.2±1.2 kg/d) were assigned to four diets in a randomized complete block design. Blocks were days in milk (B1 = 60 to 90 days, B2 = 120 to 150 days, B3 = 200 to 230 days). The diets contained FEW at 0, 25, 35 and 45% of DM in a total mixed ration. The inclusion levels were as recommended by Sunato *et al*. (2012) who used fresh ruzi grass (Brachiaria ruziziensis) as the roughage source. All diets were formulated to contain 14.5% CP and 67.5% TDN (Table 1) using the KCF 2006 software of Pattarajinda and Duangjindam (2006). Data were collected for 74 days after a 14 days adjustment period.

**Measurements, sample collection and chemical analysis:** Ethanol waste was fermented by first preparing a mixture of commercial dry yeast (S. cerevisiae) and sugar (20 g each in 1,000 ml of clean water). The mixture was activated by incubating at 30°C for 1 h before use (Khampa *et al*., 2009; Khampa *et al*., 2011). The activated mixture was added to the ethanol waste at a rate of 25% (v/w) plus 2% urea and 6% sugar (Sunato *et al*., 2012). After packing in black plastic bags (30 kg/bag), fermentation was completed at room temperature in 15 days before using FEW as a feedstuff. Weekly composites of TMR and orts were collected from daily samples of about 0.5 kg and stored at -20°C. TMR diet samples were dried for 48 h at 60°C and ground to pass through a 1 mm screen using a Wiley Mill (Thomas-Wiley, Philadelphia, PA) and analyzed for DM, CP, ether extract and ash (AOAC, 1985) and for NDF and ADF (Van Soest *et al*., 1991). Cows were housed individually in pens (2 x 3 m) with free access to water. Cows were offered TMR diet for 2 times a day (8.00 am and 4 pm.). Feed offered and orts were measured and recorded daily during the period of the experiment to calculate feed intake. Cows were milked twice daily at 6:00 am and 3:30 pm and milk production recorded. Cows were weighed on the starting and ending date of the experimental period. Milk samples were collected and composited weekly for individual cow milk analysis for fat, protein, lactose, total solid (TS) and solid not fat (SNF) using Milkosonic S-L90 procedures. On day 64 of the experimental period, 20 g of $Cr_2O_3$ was included in all diets as an external marker. Fecal samples were collected at 6 h intervals and composited for nutrient and chromium analysis to estimate dry matter and feed digestibility according to Maynard and Loosli (1975). On day 72 blood samples were collected from the coccygeal vein at hourly intervals (0, 1, 2 and 3 h) in 10 ml of tubes. Each tube was immediately placed on ice until centrifugation at 3,500 rpm for 15 min and after centrifuging, blood serum was stored at -20°C before being analyzed for blood glucose and blood urea nitrogen using an automated chemistry analyzer (HITACHI 912). Also, on day 74, rumen fluid was collected at hourly intervals (0, 1, 2 and 3 h) by a suction tube and ruminal fluid samples were obtained by straining ruminal contents through 2 layers of cheesecloth. The pH was immediately determined by using a pH meter (Electrochemical Analyzer, Consort model C933P) and then preserved by the addition of 5 ml of $1MH_2SO_4$ solution to 50 ml of rumen fluid and stored at -20°C. Prior to analysis, the ruminal fluid samples were thawed and centrifuged at 3,500 rpm for 15 min at 4°C. The supernatants were collected to determine ammonia nitrogen ($NH_3$-N) using the Kjeldahl method (Bremner and Keeney, 1965). The VFA were analyzed using an HPLC Instrument (Controller Water Model 600E with 484 UV detector) according to Zinn and Owens (1986).

**Statistical analysis:** Response variables were analyzed by GLM procedures of SAS (1996) using a randomized complete block design. Blocks were days in milk. Significance of differences in mean values among dietary treatments were reported with p-values of the F test. Means were compared by Duncan's New Multiple Range Test, with level of significance set at $p<0.05$.

## RESULTS

**Nutrient composition of diets:** The nutrient composition of ethanol waste consisted of 16.8% DM, 6.1% CP, 48.6% NDF, 34.2% ADF, 1.0% ether extract, 10.8% ash and 4.2 pH. After fermentation with yeast, FEW was found to contain 15.5% DM, 32.9% CP, 45.3% NDF, 31.7% ADF, 1.2% ether extract, 11.3% ash and a pH of 3.4, indicating that FEW could be used as an optional feedstuff even it was quite high in moisture percentage. As the level of FEW increased in the four TMR diets, DM tended to decrease while NDF and ADF increased. The CP, ether extract and ash were not affected (Table 2).

**Dry matter intake and body weight changes:** The DMI of dairy cows decreased when level of FEW increased in the four TMR diets (14.2, 15.4, 11.4 and 7.8 kg/d, respectively). The DMI per body weight (3.5, 3.9, 2.9 and 1.9%, respectively) and body weight$^{0.75}$ (2.6, 2.9, 2.4 and 2.0%, respectively) was greatest when FEW level was not more than 25% ($p<0.01$) of the diet (Table 3). For body weight parameters, the average initial weight was 405.4 kg and the average final weight was 398.2 kg. The increasing levels of FEW affected body weight changes, cows lost body weight when FEW was fed more than 25% of the diet ($p<0.01$; Table 3)

**Nutrient digestibility:** The level of FEW in the diets did not affect the apparent digestibility of ether extract, NDF and ADF (74.3, 70.7 and 70.3%, respectively). Apparent

Table 1: Formulation and calculated nutrient composition (%, DM) for increasing FEW in TMR diets

| | Level of FEW | | | |
|---|---|---|---|---|
| Ingredients | 0 | 25 | 35 | 45 |
| Ruzi grass | 35.0 | 38.6 | 32.0 | 25.3 |
| Cassava chip | 37.7 | 28.0 | 28.0 | 28.0 |
| Palm meal | 8.5 | 0.0 | 0.0 | 0.0 |
| Soybean meal | 16.3 | 6.23 | 3.06 | 0.0 |
| FEW | 0 | 25.0 | 35.0 | 45.0 |
| Mineral mix | 1.0 | 1.0 | 1.0 | 1.0 |
| Salt | 0.5 | 0.5 | 0.5 | 0.5 |
| Urea | 1.0 | 0.67 | 0.44 | 0.2 |
| **Nutrient composition (%)** | | | | |
| DM | 49.8 | 28.7 | 25.8 | 23.5 |
| CP | 14.5 | 14.5 | 14.5 | 14.5 |
| TDN | 69.2 | 66.0 | 66.0 | 66.0 |
| NDF | 35.4 | 41.0 | 40.6 | 40.3 |
| ADF | 20.6 | 24.1 | 24.3 | 24.3 |
| Ether extract | 1.3 | 1.1 | 1.1 | 1.1 |

FEW: Yeast fermented ethanol waste, DM: Dry matter, CP: Crude protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber

Table 2: Nutrient analysis of TMR diets with differing levels of FEW (%DM basis)

| | | | Level of FEW | | | |
|---|---|---|---|---|---|---|
| Item | FEW | EW | 0 | 25.0% | 35.0% | 45.0% |
| DM | 16.8 | 15.5 | 53.6 | 35.5 | 29.4 | 21.8 |
| CP | 6.1 | 32.9 | 14.4 | 14.7 | 15.0 | 15.5 |
| Ether extract | 1.0 | 1.2 | 1.2 | 1.1 | 1.1 | 1.1 |
| NDF | 48.6 | 45.3 | 51.7 | 52.4 | 54.2 | 54.4 |
| ADF | 34.2 | 31.7 | 35.4 | 35.6 | 37.3 | 38.6 |
| ash | 10.8 | 11.3 | 10.6 | 10.5 | 10.2 | 10.7 |
| pH | 4.2 | 3.4 | | | | |

EW: Ethanol waste, FEW: Yeast fermented ethanol waste, DM: Dry matter, CP: Crude protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber

digestibility of DM and CP tended to decrease when FEW was included at 45% of diet. The digestibility of DM (75.6, 74.1, 69.1 and 63.5%, respectively; p = 0.13) and digestibility of CP were (78.9, 81.9, 80.2 and 67.5%, respectively; p = 0.09) (Table 3).

**Ruminal pH:** Ruminal pH was not affected by FEW in any of the treatments, either before or after feeding (p>0.10). The average rumen pH levels at 0, 1, 2 and 3 h after feeding were 6.8, 6.9, 7.0 and 6.6, respectively (Table 3).

**Ruminal $NH_3$-N:** Ruminal $NH_3$-N was affected by the proportion of FEW in the diets. Ruminal $NH_3$-N before feeding was 8.40, 10.7, 12.6 and 13.4 mg% at 0, 1, 2 and 3 hours, respectively (p<0.05), but these differences were not significant 1 to 3 hours after feeding. The average $NH_3$-N levels in the rumen at 0, 1, 2 and 3 h after feeding were 19.0, 17.2, 18.0 and 17.9 mg%, respectively) (Table 3).

**Volatile fatty acid production in the rumen:** An inverse relationship was observed between total VFA and levels of FEW in diets. As levels of FEW increased in the diets the total ruminal VFA production decreased (110.9, 100.9, 105.8 and 94.0 mmol/l, respectively; p<0.05).

Table 3: Dry matter intake, body weight (BW) change, ruminal pH, $NH_3$-nitrogen and VFA of lactating dairy cows fed differing levels of FEW in TMR diets

| | Level of FEW | | | | | |
|---|---|---|---|---|---|---|
| Item | 0 | 25.0% | 35.0% | 45.0% | SEM | p-values |
| **DMI** | | | | | | |
| kg/d | 14.2$^{a}$ | 15.4$^{a}$ | 11.4$^{b}$ | 7.8$^{c}$ | 0.33 | <0.01 |
| BW (%) | 3.5$^{ab}$ | 3.9$^{a}$ | 2.9$^{b}$ | 1.9$^{c}$ | 0.13 | <0.01 |
| $BW^{0.75}$ (%) | 2.6$^{ab}$ | 2.9$^{a}$ | 2.4$^{b}$ | 2.0$^{c}$ | 0.05 | <0.01 |
| Initial BW, (kg) | 409.2 | 401.0 | 424.2 | 387.1 | 10.95 | 0.70 |
| Final BW, (kg) | 432.8 | 407.1 | 406.4 | 346.5 | 9.73 | 0.09 |
| BW Change, (kg) | 23.7$^{a}$ | 6.2$^{a}$ | -17.8$^{b}$ | -40.6$^{c}$ | 3.19 | <0.01 |
| **Digestion (%)** | | | | | | |
| DM | 75.6 | 74.1 | 69.1 | 63.5 | 1.65 | 0.13 |
| CP | 78.9 | 81.9 | 80.2 | 67.5 | 1.77 | 0.09 |
| Ether extract | 79.8 | 74.4 | 70.9 | 72.2 | 1.50 | 0.27 |
| NDF | 74.9 | 70.9 | 70.6 | 66.2 | 1.91 | 0.51 |
| ADF | 74.7 | 71.1 | 70.5 | 64.9 | 1.62 | 0.29 |
| **Ruminal pH** | | | | | | |
| 0-h pre feeding | 7.1 | 7.0 | 7.2 | 6.9 | 0.08 | 0.62 |
| 1-h post feeding | 7.0 | 7.0 | 7.0 | 6.6 | 0.07 | 0.27 |
| 2-h post feeding | 6.8 | 6.8 | 7.2 | 6.6 | 0.07 | 0.11 |
| 3-h post feeding | 6.7 | 6.8 | 7.0 | 6.7 | 0.09 | 0.51 |
| 1-3 h post feeding | 6.8 | 6.9 | 7.0 | 6.6 | 0.06 | 0.19 |
| **$NH_3$-N, (mg %)** | | | | | | |
| 0-h pre feeding | 8.40$^{b}$ | 10.7$^{ab}$ | 12.6$^{a}$ | 13.4$^{a}$ | 0.48 | 0.04 |
| 1-h post feeding | 14.6 | 18.2 | 16.8 | 17.8 | 1.03 | 0.66 |
| 2-h post feeding | 22.4 | 17.7 | 19.1 | 19.9 | 0.79 | 0.29 |
| 3-h post feeding | 20.0 | 15.8 | 18.2 | 16.2 | 1.12 | 0.56 |
| 1-3 h post feeding | 19.0 | 17.2 | 18.0 | 17.9 | 0.34 | 0.40 |
| Total VFA, mmol/L | 110.9$^{a}$ | 100.9$^{bc}$ | 105.8$^{ab}$ | 94.0$^{c}$ | 1.26 | 0.02 |
| **Molar proportion of VFA (mol/100 mol)** | | | | | | |
| Acetate (C2) | 51.7 | 54.4 | 54.7 | 52.9 | 1.00 | 0.12 |
| Propionate (C3) | 30.8 | 26.4 | 28.2 | 27.4 | 0.94 | 0.40 |
| Butyrate (C4) | 17.4 | 19.3 | 17.2 | 19.7 | 0.63 | 0.52 |
| C2/ C3 | 1.8 | 2.1 | 1.9 | 1.9 | 0.08 | 0.48 |

Values with different letters in the same row differ significantly (p<0.05) FEW: Yeast fermented ethanol waste, DMI: Dry matter intake, BW: Body weight, $BW^{0.75}$: Metabolic of the body weight, DM: Dry matter, CP: Crude protein, NDF: Neutral detergent fiber, ADF: Acid detergent fiber, $NH_3$-N: Ammonia nitrogen, VFA: volatile fatty acids

Table 4: Blood glucose and blood urea nitrogen of lactating dairy cows fed differing levels of FEW in TMR diets

| | Level of FEW | | | | | |
|---|---|---|---|---|---|---|
| Item | 0 | 25.0% | 35.0% | 45.0% | SEM | p-values |
| **Glucose, (mg/dL)** | | | | | | |
| 0-h pre feeding | 62.7$^{b}$ | 62.7$^{b}$ | 63.0$^{b}$ | 68.3$^{a}$ | 0.40 | <0.01 |
| 1-h post feeding | 67.7 | 69.3 | 67.0 | 61.3 | 1.30 | 0.25 |
| 2-h post feeding | 70.7 | 68.3 | 68.3 | 64.0 | 1.38 | 0.45 |
| 3-h post feeding | 71.7 | 68.3 | 70.3 | 65.3 | 1.69 | 0.60 |
| 1-3 h post feeding | 70.0 | 68.7 | 68.6 | 63.6 | 1.24 | 0.36 |
| **BUN, (mg/dL)** | | | | | | |
| 0-h pre feeding | 9.7 | 20.3 | 18.0 | 17.7 | 1.37 | 0.13 |
| 1-h post feeding | 12.3 | 21.0 | 19.3 | 18.7 | 1.44 | 0.26 |
| 2-h post feeding | 13.0 | 22.0 | 20.7 | 19.7 | 1.45 | 0.23 |
| 3-h post feeding | 12.3 | 22.0 | 21.0 | 20.3 | 1.32 | 0.13 |
| 1-3 h post feeding | 12.6 | 21.7 | 20.3 | 19.6 | 2.40 | 0.20 |

Values with different letters in the same row differ significantly (p<0.05) FEW: Yeast fermented ethanol waste, BUN: Blood urea nitrogen

Acetate tended to increase when FEW was up to 35% of the diets and then decreased at the 45% level (51.7, 54.4, 54.7 and 52.9%, respectively; p = 0.12). The level of FEW in the diets did not affect propionate concentration (30.8, 26.4, 28.2 and 27.4%, respectively),

Table 5: Milk production and milk composition of lactating dairy cows fed differing levels of FEW in TMR diets

| | ---------- Level of FEW ---------- | | | | | |
|---|---|---|---|---|---|---|
| Item | 0 | 25.0% | 35.0% | 45.0% | SEM | p-values |
| Milk yield, (kg/d) | 10.8[a] | 11.1[a] | 9.9[ab] | 8.1[b] | 0.28 | 0.04 |
| Milk 4% FCM, (kg/d) | 11.6 | 11.7 | 11.0 | 8.9 | 0.28 | 0.07 |
| Milk Fat, (%) | 4.6 | 4.4 | 4.8 | 3.6 | 0.18 | 0.22 |
| Milk Protein, (%) | 3.0 | 2.9 | 2.9 | 2.9 | 0.04 | 0.74 |
| Lactose, (%) | 4.1 | 4.0 | 3.9 | 3.9 | 0.05 | 0.40 |
| Milk TS, (%) | 12.2 | 11.9 | 12.2 | 11.1 | 0.21 | 0.26 |
| Milk SNF, (%) | 7.8 | 7.6 | 7.4 | 7.5 | 0.09 | 0.58 |

Values with different letters in the same row differ significantly ($p<0.05$). FEW: Yeast fermented ethanol waste, 4% FCM: Milk yield (0.4+0.15% Fat), TS: Total solids, SNF: Solids not fat

butyrate concentration (17.4, 19.3, 17.2 and 19.7%, respectively) and C2:C3 ratio (1.8, 2.1, 1.9 and 1.9, respectively) in rumen (Table 3).

**Blood glucose and Blood urea nitrogen (BUN):** There were no differences in Blood glucose concentrations (BG) before and after feeding, at 1, 2 and 3 h post feeding, BG concentrations for the treatments were 70.0, 68.7, 68.6 and 63.6 mg/dL, respectively (Table 4). Blood urea nitrogen concentrations (BUN) was not affected ($p = 0.13$). The average BUN level at 1, 2 and 3 h post feeding were 12.6, 21.7, 20.3 and 19.6 mg/dL, respectively. An increasing BUN concentration was observed at 1 h ($p = 0.26$), 2 h ($p = 0.23$) and 3 h ($p = 0.13$) after feeding, with nearly a two-fold difference (Table 4) and BUN concentration was highest at 3 h after feeding as evidenced in all treatments.

**Milk production and milk composition:** There was increase in milk yield in cows receiving 25% of FEW in the diet but decreased when FEW exceeded 35% (10.8, 11.1, 9.9 and 8.1 kg/d, respectively; $p<0.05$). The levels of FEW in the diets did not affect milk composition, average milk fat, protein, lactose, TS and SNF (4.4, 2.9, 4.0, 11.8 and 7.6%, respectively; $p>0.05$) which were within this standard milk composition (Table 5).

## DISCUSSION

There were reductions in DMI with increased levels of FEW in current study which might be due to high moisture content in FEW and TMR (Miller-Cushon and DeVries, 2009; Felton and Devries, 2010). Lahr *et al.* (1983) reported that DMI decreased from 22.3 to 19.4 kg/d, when the moisture content of diet increased from 22 to 60%. Also, as summarized in NRC (2001), DMI decreases by 0.02% of BW for each 1% increase in moisture content when it is greater than 50% of the diet. Besides moisture, bulk density and gut distention may affect DMI (Dado and Allen, 1995; Ruiz *et al.*, 1995; Eastridge, 2006).

There were decreasing trend in body weight with increasing levels of FEW, because the amount of feed intake was affected by moisture and NDF content in diets (Tjardes *et al.*, 2002). The DMI in animals supply nutrients for production, improving body weight and maintenance.

The DM digestibility in this study is contrary to the Oba and Allen (2003) and Krause *et al.* (2002) reported that DM digestibility increased at higher level of fermented carbohydrate inclusion in the diet. This could possibly be due to ingesta passing at a rapid rate, caused by elevated moisture content and less bulk density of the diet. The FEW is a composite of extra fermentable protein, carbohydrate and moisture as it is fermented for 15 days before feeding and supplemental FEW in the right proportion might promote microbial protein synthesis and maintain rumen pH and rumen function (Knowlton, 2000).

Ruminal pH tended to decrease at 2 h after feeding and this may be due to more moisture content and lactic acid production through fermentation of the diet. However, the pH remained quite stable (near neutral 7.0) 3 h after feeding which is ideal for microbial protein synthesis in the rumen. Also, TMR feeding and the readily degradable protein in the diets produced more ruminal $NH_3$, thus helping to maintain ruminal pH. Consequently, FEW in rations had the dual positive effects of improving protein degradation and maintaining pH. Schwartzkopf-Genswein *et al.* (2003) and Bach *et al.* (2005) reported that ruminal pH above 6.5 would promote the growth rate of ruminal microorganism and promote protease activity. The $NH_3$-N production is known to reach peak at 2 h after feeding with a concentration greater than that of pre-feeding (Geerts *et al.*, 2004). NRC (2001) reported that ruminal $NH_3$-N would reach the maximum level within 1.5 to 2.0 hour post-feeding due to protein degradation and that a range of 15 to 30 mg% was obtained by Wanapat and Pimpa (1999).

The result indicate that FEW in diets could result in a more rapid rate of passage than the control diet since FEW diets had more fermentable nutrients and a greater moisture content. Consequently, diet containing higher levels of FEW lowered digestibility of NDF and ADF. These levels were within the normal range of 70 to 130 mmol/l (France and Siddons, 1993). In general, propionate concentrations increased but acetate and butyrate concentration decreased as the concentrate to roughage ratio (C:R ratio) increased. In this study lower concentration of acetate was found as compared to propionate and butyrate which caused the C2:C3 ratio to decrease. Ishler *et al.* (1996) reported that changing the C:R ratio from 50:50 to 80:20 changed the acetate, propionate and butyrate concentration ratios from 65.3-18.4, 10.4-53.6 and 30.6-10.7, respectively. Similarly, Sutton *et al.* (2003) confirmed that a change from 60% to 90% concentrates in total mixed ration doubled the propionate concentrations and decreased acetate and butyrate concentrations.

Blood glucose concentrations (BG) in this study were within the normal range (43.2-68.4 mg/dL) as shown by

Mudron *et al*. (2005). The data indicated that all FEW diets had sufficient rumen fermentable carbohydrates compared to the more soluble carbohydrates of control diet (0% FEW).

An increase in Blood urea nitrogen concentrations (BUN), reflects the increased CP digestion with increasing levels of FEW diets. Furthermore, the protein in FEW is known to have a relatively high rate of rumen degradability, thus explaining these differences. The BUN was highest at 3 h after feeding as evidenced in all treatments, according to Gustafson and Palmquist (1992) the ruminal $NH_3$-N was absorbed into the blood stream within 1.5-2 hour. Also indicating that the BUN levels over 20 mg/dL in this study were mostly due to rapid ruminal degradation of protein in FEW diets. Kohn *et al*. (2005) reported that the normal levels of BUN ranged from 4 to 25 mg/dL.

Milk production in this study decreased with increasing level of FEW in diets, because of the reduced DMI which is positively related to milk production. The decreasing DMI at FEW levels above 25% reduced the energy intake and increased the rate of passage of ingesta due to high moisture content (Martin and Sauvant, 2002; Kendall *et al*., 2009). There were no affect on milk composition, VFA production and carbohydrate digestion in dairy cows fed FEW.

**Conclusion:** Improving the nutrient value of ethanol waste by fermenting with yeast for 15 days could improve protein content and fermentable carbohydrates. Feeding up to 25% FEW in TMR diets has the potential to increase DMI and milk production. Supplementation with more than 25% levels of FEW; decreased the DMI and TMR digestibility due to increased moisture content. However, supplemental FEW did not affect BG, BUN, VFA, ruminal pH or $NH_3$-N production. Fermenting with yeast is the best treatment for improving ethanol waste as a feedstuff and increasing its potential use as a protein source for lactating dairy cattle. However, controlling moisture content is important for successful utilization of FEW as a feedstuff.

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