Acta Scientiarum. Animal Sciences
ISSN: 1807-8672
Editora da Universidade Estadual de Maringá - EDUEM

Reza Gerimipour, Ali; Azin, Mehrdad; Reza Sanjabi, Mohammad
The effect of vinasse on the growth performance and feed digestibility of Holstein male calves
Acta Scientiarum. Animal Sciences, vol. 41, e45666, 2019
Editora da Universidade Estadual de Maringá - EDUEM

DOI: https://doi.org/10.4025/actascianimsci.v41i1.45666

Acta Scientiarum
http://periodicos.uem.br/ojs/acta
ISSN on-line: 1807-8672
Doi: 10.4025/actascianimsci.v41i1.45666

RUMINANTS NUTRITION

The effect of vinasse on the growth performance and feed digestibility of Holstein male calves

Ali Reza Gerimipour[1], Mehrdad Azin[1] and Mohammad Reza Sanjabi[2*]

[1]Biotechnology Department, Iranian Research Organization for Science and Technology, Tehran, Iran. [2]Agriculture Research Institute, Iranian Research Organization for Science & Technology, Sh. Ehsani Rad St., Enqelab St., Parsa Sq., Ahmadabad Mostoufi Rd., Azadegan Highway, 33535-111, Tehran, Iran. *Author for correspondence. E-mail: msanjabii@gmail.com

ABSTRACT. Industrial effluents of alcohol production are problematic for the environment. One of the proposed solutions to the reduction of harmful effects of the wastewater is to convert it into vinasse. The purpose of the present study is to investigate effects of rations with different levels of vinasse as a source of protein in the diet of 28 male Holstein calves, previously health checked, with an average initial weight of 300 ± 22.56 kg and the maximum similarity of weight and age. The calves were divided randomly into four treatments varied in vinasse (substituted for cottonseed meal): 0 (control), 5, 10 and 15 % with seven replications and each group was given its own specific diet for 110 days. Growth performance, nutrient digestibility and feed intake were examined. Final body weight, average daily weight gain and dry matter intake were significantly higher in calves fed diet with 10% vinasse compared to other treatments ($p < 0.05$). Rumen pH significantly increased in treatment four (15% vinasse) ($p < 0.05$). Crude fat, NDF, non-fiber carbohydrate (NFC) and dry matter digestibility didn't show any significant difference between treatments ($p > 0.05$), but treatments with 10 and 15% vinasse significantly increased organic matter andcrude protein digestibility compared with control ($p < 0.05$). Totally substituting cottonseed meal with 10% vainasse in calves diet improved body weight gain and organic matter digestibility without any significant effects on rumen pH.

Keywords: Daily Gain; feed Intake; non-protein nitrogen; by-product.

Received on December 4, 2018.
Accepted on April 11, 2019.

Introduction

The alcohol industry has developed quickly which lead to increase the hazards to the environment. One of the proposed solutions to reducing their harmful effects is to convert the wastewater into a substance called vinasse. Vinnase is a dark red liquid with an acidic pH around 4.5, high viscosity and about 65% dry matter. This substance can be stored for unlimited time in a warehouse and has easy trasnport. Vinnase is obtained from molasses (Hannon & Trenkle, 1990), and since a remarkable proportion of the molasses is used in alcohol production, its non-sugar components and minerals accumulate in vinasse. It is low in sugar but its amount of protein and minerals is relatively high. Vinasse is a substance of low value and has many applications, including cosmetics, biogas, fertilizer and animal feed as a source of nutrients and minerals (Leontowicz et al., 1994; Lopez-Campos et al., 2011; Stemme, Gerdes, Harms, & Kamphues, 2005; Yalcin, Eltan, Karsli, & Yalcin, 2010). Due to the constraint of resources in fodder and grain production in some countries and the high cost of using grains such as oat, corn, soybean and cottonseed as animal feed, vinasse is a cheap and useful substitute for animal feed. Furthermore, high viscosity of vinasse facilitates the attachment of various components of feed which prevents selective eating of the animal and reduces the dust of mash diets too. Vinnase also has an essential role in production of pellet-form feed.

As vinasse contains the decomposed remains of yeasts and plenty of soluble protein which is mostly non-protein nitrogen (NPN) and high amount of minerals which are highly available, is used vastly for livestock feeding specially ruminants (Baile & Forbes, 1974; Leontowicz et al., 1994). It also has plenty of vitamin B and minerals that can be used for animal feed (Davis et al., 2005). The desirable profile of amino acids in vinasse allows its usage for a wide variety of species of animals and can be useful in activating their immune system. The percentage of minerals especially elements such as iron, calcium, and phosphorus in fermented vinasse is suitable for animals (Lopez-Campos et al., 2011). The calcium in vinasse can play a significant role in strengthening bones, teeth, blood clotting and transmission of nerve impulses in livestock.

The history of using vinasse for feeding livestock goes back to the 1960s in some European countries (Gallo, Gip Ospiua, & Santos, 2008; Hammond, 1995). It has been reported that vinasse can notably improve the digestibility and efficiency of feed for cattle (Gallo et al., 2008; Konan, Antolikova, & Husar, 1987; Leontowicz et al., 1994). Vadivel et al. (2014) suggested that the sugar and starch compounds of vinasse can increase feed intake and the survival rate of animals at the time of food shortage. Much of its crude protein in combination with amide compounds is palatable to livestock and can be quickly used by rumen microbes in microbial protein synthesis. It has been showed that the digestibility of vinasse organic matter in cattle is approximately 70% (Stemme et al., 2005). So the purpose of this study was to substituting cottonseed meal with different percentages of vinasse obtained from the fermentation of molasses as a source of microbial protein in the diet of fattening Holstein bull calves.

Material and methods

Chemical analysis of vinasse

Sampling was done at 3 different times from various parts of produced vinasse in Bidestan alcohol factory and samples were thoroughly mixed and transferred to a laboratory after 35 days to determine its nutrients and microbial changes. The dry matter was measured at a temperature of 55°C for two days in the oven. The ingredients of the vinasse was analyzed as crude fat, crude protein, crude fiber, minerals, ash and heavy metals according to Association Official Analytical Chemist (AOAC, 2005).

Experimental design

In this study, 28 male Holstein calves, previously health checked, with an average initial weight of 300 ± 22.56 kg and the maximum similarity of weight and age were selected. The calves were divided randomly into four groups of seven and each group was given regular diet plus 0% vinasse (treatment 1, control), 5% vinasse (treatment 2), 10% (treatment 3) and 15% (treatment 4) as replacement with cottonseed meal respectively. The duration of the study was 110 days, with 20 days considered as the adaptation period to the experiment.

The experimental diets and feeding strategy

Food rations were adjusted using the software National Research Council (NRC, 2001) (70% concentrate based on dry matter) in four categories which had the same energy and protein.

The diets were selected considering the composition, nutritional value, and 5 kg of daily dry matter for each cattle (Table 1).

Table 1. Composition of the basal diet.

Ingredients, %	Vinasse level (%)			
	0	5	10	15
Vinasse Level	0	5	10	15
Alfalfa	7.7	7.7	7.7	7.7
Corn Silage	46.15	46.15	46.15	46.15
Corn	15.4	15.4	15.4	15.4
Barley	15.4	15.4	15.4	15.4
Beet Pulp	2.3	2.3	2.3	2.3
Fat Powder	1.53	1.53	1.53	1.53
Cotton seed meal	3.84	2.61	1.38	0
Magnesium Oxide	0.46	0.46	0.46	0.46
Calcium Carbonate	0.23	0.23	0.23	0.23
Sodium Bicarbonate	0.23	0.23	0.23	0.23
Salt (g)	0.07	0.07	0.07	0.07
Vitamin Supplement*	0.07	0.07	0.07	0.07
Trace Mineral	0.02	0.02	0.02	0.02
Molasses	1.53	1.53	1.53	1.53
Wheat Bran	3.84	3.84	3.84	3.84
Soybean Meal	0.77	0.77	0.77	0.77

*Each kilogram of vitamin supplement contained vitamin A (600,000 IU), vitamin D (200,000 IU), vitamin E (200mg), antioxidants (2500 mg), calcium (195 g), phosphorus (80 g), magnesium (21,000 mg), manganese (2200 mg), iron (3000 mg), copper (300 mg), zinc (300 mg) cobalt (100 mg), iodine (120 mg), and selenium (1.1 mg).

Measuring feed intake and body weight changes

Daily feed intake has been measured and controlled and body gain measured monthly.

Measuring the apparent digestibility of nutrients

Apparent digestibility of nutrients was determined using acid-insoluble ash according to (Van Keulen & Young, 1977) method. Dried samples of feces and feed were put in a furnace for 6 hours at 550°C temperature to obtain ash. Lumbar puncture was done on a monthly basis to determine and control rumen pH. The chemical composition of meat samples was determined using AOAC (2005). The crude fat of meat samples, ash and crude protein content were measured. Crude fat was estimated using petroleum ether in a Soxhlet apparatus. Briefly, crude sample was folded in a filter paper, placed in extraction thimble and cotton wools placed on top. Then about 300 cm^3 of 60–80C° petroleum ether was added into the flask. The extraction was last three hrs. The samples were air-dried and then oven dried at 80C°. The extractible fat was then calculated as percentage crude fat (%). For crude protein, nitrogen was determined by Kjeldahl method. Sample's ash content were measured using AOAC (2005), in 86°C oven for 24h and 600°C furnace.

Data analysis

Statistical analysis of the collected data was conducted using GLM method of the statistical software of the Statistical Analysis System (SAS, 2004). Before analyzing the data, the mean comparison was performed using Duncan's multiple range Test ($p \leq 0.05$). The mixed procedure was used to analyze data with replication such as parameters of the rumen, average daily gain, and feed intake. The results were statistically analyzed by using PROC GLM (General Linear Models) in the SAS software. Statistical analysis for achieving linear, quadratic and cubic contrasts was carried out by observing the response types of the variables to the assessed diets. Regression analyses were performed to achieve estimates of the assessed responses for the levels of vinasse not used in the experiment. PROC REG in the SAS software was used for this estimative.

Results

The chemical composition of vinasse

The chemical composition of vinasse used in this study is shown in Table 2. Although the ash content in vinasse is high, from mineral point of view it is rich of calcium (17.5 g kg^{-1}), sodium(19.2 g kg^{-1}) and high potassium amount.

Table 2. Chemical compositions and minerals of the vinasse.

Parameter	Content
Dry matter (%)	64.6
Ash (%)	14.8
Crude fat (%)	0.15
Crude protein (%)	20
Calcium (g kg^{-1})	17.5
Phosphorus (g kg^{-1})	0.8
Sodium (g kg^{-1})	19.2
Potassium (g kg^{-1})	64
Magnesium (g kg^{-1})	2.3
Manganese (g kg^{-1})	1.3
Zinc (g kg^{-1})	0.028
Iron (g kg^{-1})	1.4
Copper (g kg^{-1})	0.0021

The level of heavy metals in vinasse is low. In comparison to standard values, it is within the allowable range (Table 3).

Table 3. Analysis of heavy metals in vinasse.

Heavy Metal	Cr (ppm)	Ni (ppm)	Cd (ppb)	Hg (ppb)	Pb (ppm)	As (ppm)	Fe (ppm)
Content	1.21±0.18	4.72±1.55	119.34±22.14	8.93±2.45	3.64±0.78	0.71±.12	644.8±130.12

Weight gain and feed intake

There was a difference between the averages final weight of calves fed treatment 3 compared to other treatments ($p \leqslant 0.05$). Increasing the amount of vinasse to 10% of dry matter ration, increased the final weight and average daily weight gain of calves. However, no difference was observed between the treatments 2 and 4 (Table 4).

Average Daily Weight Gain increased by 4.06 kg day^{-1} for every 10 g of vinasse of concentrate, as demonstrated by the regression equation: $\mathrm{ADWG(kg)} = 112.7 + 4.06\mathrm{X} - 0.219$, $p < 0.05$) by cubic contrast, where: x = the inclusion level of vinasse in the concentrate.

The effects of vinasse levels on dry matter intake of calves during the experiment (Table 4). Dry matter intake increased significantly in treatment 3 (10% vinasse) compared with control and 15% vinasse. Vinasse level showed quadratic effects on dry matter intake and it increased by 0.48 kg for every 10 g of vinasse in ration ($\mathrm{Y} = 7.99 + 0.48\mathrm{X} - 0.039$, $p < 0.05$).

Rumen pH

As it can be seen in Table 5, there was no remarkable difference in rumen pH among treatments, but rumen pH decreased notably during the third and fourth periods. However, rumen pH increased by increasing the levels of vinasse. In the experiment period, a difference was seen between treatments, whereas rumen pH significantly increased in treatment 4 (15% vinasse).

Apparent digestibility of diets

The average apparent digestibility (%) of diets has been reported in Table 6. Crude fat (CF), Non-fibrous carbohydrates (NFC), NDF and dry matter (DM) digestibility didn't affect significantly by dietary treatments ($p \geqslant 0.05$). Crude protein and organic matter digestibility increased significantly compared with control ($p < 0.05$). Crude protein digestibility increased by quadratic contrast ($\mathrm{CP} = 70.44 + 0.45\mathrm{X} - 0.007$, $R^2 = 0.41$).

Table 4. Weight gain and feed intake of calves in different rations.

Parameter	Vinasse (%) 0		5		10		15
Initial Weight (kg)	308.26±22.65		299.85±20.39		317.81±24.42		321.58±20.73
Final Weight (kg)	409.95±21.14^b		412.34±19.11^b		472.15±20.19^a		434.13±18.43^b
Average Daily Weight Gain (kg)	1.15±0.06^b		1.17±0.07^b		1.37±0.09^a		1.03±0.09^b
Dry Matter Intake	8.23±0.24^b		8.74±0.51^{ab}		9.49±0.27^a		7.14±0.35^b
	Linear		Quadratic		Qubic		
	Probability	F ratio	Probability	F ratio	Probability	F ratio	R^2
ADWG	0.5328	0.40	0.4337	0.64	0.0525	4.25	0.60
DM	0.0574	4.07	0.0042	10.40	0.0952	3.07	0.67

^{a,b,c}Row means with different superscripts differ significantly at ($p < 0.05$). $\mathrm{ADWG(kg)} = 112.7 + 4.06\mathrm{X} - 0.219$, $\mathrm{DM} = 7.99 + 0.48 - 0.039$.

Table 5. Rumen pH changes in different treatments.

Parameter	Vinasse (%) 0		5		10	15	
Rumen pH	6.92±0.05^b		7.02±0.09^b		7.08±0.05^b	7.21±0.06^a	
Linear		Quadratic		Qubic			
Probability	F ratio	Probability	F ratio	Probability	F ratio	R^2	
0.0180	6.64	0.0621	3.90	0.0752	3.52	0.55	

^{a,b,c}Row means with different superscripts differ significantly at ($p < 0.05$). $\mathrm{Y} = 6.96 + 0.018\mathrm{X} - 0.0004$.

Discussion

According to the results of present research and what Konan et al. (1987) reported, adding vinasse to the diet of young bulls, increased feed intake and consequently daily weight gain got higher in calves. They attribute this to the palatability of vinasse due to its amide compounds. In the first study, increasing the level of vinasse up to 5% of the diet increased the feed intake and final weight. Sampaio, Oliveira, Kronka, and Oliveira (1989) reported that adding vinasse up to 10% of dry matter to a diet consisting 50 percent concentrate and 50 percent fodder increased the final weight. In this experiment due to the lack of difference between the initial weight of calves fed different diets and also the indifference among the energy of all three diets, the increased final weight can be attributed to varying levels of vinasse in various treatments.

Table 6. The effects of dietary treatments on nutrients digestibility.

Nutrient	Treatments (% vinasse)				Contrasts						
					Linear		Quadratic		Cubic		
	0	5	10	15	P	F	P	F	P	F	R^2
Crude fat	65.55±3.59	63.79±3.71	68.12±3.31	68.74±1.87	0.2234	1.58	0.308	1.09	0.605	0.28	0.11
Crude protein	70.31±2.94 ^{b}	72.37±2.28 ^{ab}	75.45±3.09 ^{a}	75.77±1.13 ^{a}	0.3222	1.03	0.0733	3.57	0.0539	4.20	0.41
NDF	45.60±2.17	47.92±3.50	46.67±2.21	49.79±2.15	0.0304	5.43	0.6788	0.18	0.8368	0.04	0.21
NFC	91.87±1.39	93.56±2.78	94.91±3.65	93.89±2.50	0.6806	0.17	0.6232	0.25	0.2757	1.26	0.29
Dry matter	73.76±1.78	75.54±2.92	77.71±2.61	78.50±1.63	<0.0001	29.25	0.5330	0.40	0.3210	1.04	0.58
OM	73.78±1.53 ^{b}	75.80±2.21^{ab}	79.31±2.93 ^{a}	79.64±1.04 ^{a}	0.0005	17.04	0.3125	1.07	0.2673	1.30	0.75

[a,b,c]Row means with different superscripts differ significantly at ($p < 0.05$). OM: Organic Matter. NDF: Neutral Detergent Fiber. NFC: Non-Fiber Carbohydrate. DM: Dry Matter. $OM = 74.82 + 0.05X + 0.017$, $DM = 73.16 + 0.5X - 0.0009$,$NFC = 91.31 + 0.98X - 0.05$, $NDF = 46.68 + 0.11X - 0.01$, $CP = 70.44 + 0.45X - 0.0007$, $CP = 151.7 - 19.62X + 0.96$

Kemenade, den Hartog, Haaksma, and Verstegen (1988) obtained similar results which showed the feed intake of bull calves and broilers increases by vinasse addition and this consequently causes a greater average daily weight gain in these animals (Kemenade et al., 1988). But Maneerat, Prasanpanich, Tumwasorn, Laudadio, and Tufarelli (2015) have shown reduction in feed intake in steers fed by bassage-vinasse compared with molasses. Also, Zali, Eftekhari, Fatehi, and Ganjkhanlou (2017) have reported higher live slaughter weight in Holstein male calves fed with diet containing 5% vinasse. Since vinasse has a notable amount of soluble carbohydrates and most of its protein is combined with amides, this makes the diet significantly palatable by considering the principles of diet formulation and dietary cation and anion balance (DCAB). Additionally, vinasse has large amounts of crude protein and nitrogen compounds that can be used by microorganisms in the rumen and improve the feed digestion as well.

However, for the diet 2, there was no difference which was consistent with the results of Potter et al. (1985), Kemenade et al. (1988) and Kirchgessner and Weigand (1980). Potter et al. (1985) used vinasse at 10,5% and 15% levels of the dry matter ration fed to bulls and reported that adding vinasse up to 5% of dry matter ration has positive benefits such as increasing the voluntary intake, digestibility of feed, and daily gain of calves. The upper level of vinasse can lead to reduction of interference of Potassium metabolisms in this level of magnesium. No significant difference was observed between the average dry matter intake of calves fed diets 1 and 2. However, the feed intake in diet 2 (10% vinasse) increased which was probably due to the reduction of dry matter in the ration. In the whole period, a difference was found in the average dry matter intake among diet 4 (15% vinasse), diet 1 (0% vinasse), and diet 3 (10% vinasse). Thus, according to the results of the whole period, a difference was observed in the average dry matter intake of calves fed diets 3 compared to groups 1and 4 ($p < 0.5$). The highest intake was in diet 3 (9.49 ± 0.27 kg of dry matter intake) and the lowest intake was in diet 4 (7.14 ± 0.35 kg of dry matter intake) which wasn't consistent with the results of Kemenade et al. (1988). It is because they concluded that increasing vinasse in proportion to molasses increases the daily intake and growth rate. It should be noted that dietary compounds in the two studies are not the same and vinasse can be added up to 15% by balancing potassium level and reducing alfalfa, but in the present study, 15% level of vinasse did cause the significant reduction of feed intake during the whole period. Vinasse addition up to 10% level of dry matter increases the feed intake due to the reduction of dry matter so that cattle need to take more food to supply their required energy. Accordingly, adding vinasse to diets containing roughage, increases feed intake. Also, vinasse has an indirect impact on the microbial population of the rumen which provides minerals and microbial nitrogen that leads digestibility improve and feed intake. In the present study, increasing the level of vinasse in the diets enhanced the apparent digestibility of NDF, non-fibrous carbohydrates, and dietary dry matter but they were not significant. The reason for this may be several different mechanisms, which are effective in reducing the digestibility of fibers.

Leontowicz et al. (1994) added vinasse up to 16% of dry matter in the ration of bulls consisted of 67% fodder and 33% concentrate. Increasing levels of vinasse causes a rise in pH of the rumen which is quite similar to our results. But Fernández et al. (2009) didn't observe significant differences for ruminal liquor pH in ewes fed sugar beet pulp with different amounts of vinasse.

Since the average concentration of ammonia nitrogen is related to the produced rumen fluid ammonia, which is in good agreement with the results of pH in Table 5. Although the average dry matter intake was decreased in treatment 4, vinasse was effective on pH and concentration of ammonia nitrogen. Therefore, an increase of pH in this treatment is completely related to vinasse.

Conclusion

This study shows that increasing vinasse level of the fattening diet of the male calves up to 10% of dry matter improved average daily gain of calves, final weight, dry matter intake of calves and their rumen pH. Vinasse supplementation had a great effect on fat and dry matter digestibility, but crude protein and organic matter digestibility has improved in 10 and 15% vinasse compared with control.

References

Association Official Analytical Chemist [AOAC]. (2005). *Official Methods of Analysis* (18th ed.). Gaitherburg, MD: AOAC International.

Baile, C. A., & Forbes, J. M. (1974). Control of feed intake and regulation of energy balance in ruminants. *Physiological Reviews, 54*(1), 160-214. doi: 10.1152/physrev.1974.54.1.160

Davis, L., Jeon, Y.-J., Svenson, C., Rogers, P., Pearce, J., & Peiris, P. (2005). Evaluation of wheat stillage for ethanol production by recombinant Zymomonas mobilis. *Biomass and Bioenergy, 29*(1), 49-59. doi: 10.1016/j.biombioe.2005.02.006

Fernández, B., Bodas, R., López-Campos, Ó., Andrés, S., Mantecón, A. R., & Giráldez, F. J. (2009). Vinasse added to dried sugar beet pulp: Preference rate, voluntary intake, and digestive utilization in sheep. *Journal of Animal Science, 87*(6), 2055-2063. doi: 10.2527/jas.2008-1550

Gallo, B. J., Gip Ospiua, P. H., & Santos, V. E. (2008). Preliminary evaluation of vinasse, a distillery by-products, as a possible source of nutrients in the poultry. *Acta Agronomica, 36*(2), 207-220.

Hammond, A. C. (1995). Leucaena toxicosis and its control in ruminants. *Journal of Animal Science, 73*(5), 1487-1492. doi: 10.2527/1995.7351487x

Hannon, K., & Trenkle, A. (1990). Evaluation of condensed molasses fermentation solubles as a nonprotein nitrogen source for ruminants. *Journal of Animal Science, 68*(9), 2634-2641. doi: 10.2527/1990.6892634x

Kemenade, P., den Hartog, L. A., Haaksma, J., & Verstegen, W. M. A. (1988). Beet vinasse in pig nutrition - Effect on digestibility and production. *Mededeling. Instituut voor Rationele Suikerproduktie, 18*, 52.

Kirchgessner, M., & Weigand, E. (1980). Broiler fattening trial with vinasse and molasses in the complete diet. *Archiv fur Geflugelkunde, 44*(3), 119-123.

Konan, V., Antolikova, M., & Husar, M. (1987). Use of di stillery by products for animal feeding. *Agrochemia, 27*, 219-221.

Leontowicz, H., Krzeminski, R., Leontowicz, M., Kulasek, G., Tropilo, J., & Sobczak, E. (1994). Condensed beet molasses solubles for fattening bulls. *Journal of Animal and Feed Sciences, 3*(1), 23-31. doi: 10.22358/jafs/69815/1994

Lopez-Campos, O., Bodas, R., Prieto, N., Frutos, P., Andrés, S., & Giráldez, F. J. (2011). Vinasse added to the concentrate for fattening lambs: Intake, animal performance, and carcass and meat characteristics. *Journal of Animal Science, 89*(4), 1153-1162. doi: 10.2527/jas.2010-2977

Maneerat, W., Prasanpanich, S., Tumwasorn, S., Laudadio, V., & Tufarelli, V. (2015). Evaluating agro-industrial by-products as dietary roughage source on growth performance of fattening steers. *Saudi Journal of Biological Sciences, 22*(5), 580-584. doi: 10.1016/j.sjbs.2015.01.015

National Research Council [NRC]. (2001). *Nutrient Requirements of Dairy Cattle* (7th ed.). Washington, DC: Natlional Academy Press.

Potter, S. G., Moya, A., Henry, P. R., Palmer, A. Z., Becker, H. N., & Ammerman, C. B. (1985). Sugarcane condensed molasses solubles as a feed ingredient for finishing cattle. *Journal of Animal Science, 60*(3), 839-846. doi: 10.2527/jas1985.603839x

Sampaio, A. V. P. D., Oliveira, M. D., Kronka, S. N., & Oliveira, D. (1989). Digestibility of rations containing different levels of dry vinasse of the nutrient digestibility in bovine feeding. *Pesquisa Agropecuária Brasileira, 24*(10), 1339-1342.

Statistical Analysis System [SAS]. (2004). *SAS/STAT User guide, Version 9.1.2*. Cary, NC: SAS Institute Inc.

Stemme, K., Gerdes, B., Harms, A., & Kamphues, J. (2005). Beet-vinasse (condensed molasses solubles) as an ingredient in diets for cattle and pigs–nutritive value and limitations. *Journal of Animal Physiology and Animal Nutrition, 89*(3-6), 179-183. doi: 10.1111/j.1439-0396.2005.00554.x

Vadivel, R., Minhas, P. S., Kumar, S., Singh, Y., Nageshwar, R. D. V. K., & Nirmale, A. (2014). Significance of vinasses waste management in agriculture and environmental quality-Review. *African Journal of Agricultural Research, 9*(38), 2862-2873.

Van Keulen, J., & Young, B. A. (1977). Evaluation of Acid-Insoluble Ash as a Natural Marker in Ruminant Digestibility Studies. *Journal of Animal Science, 44*(2), 282-287.

Yalcin, S., Eltan, Ö., Karsli, M. A., & Yalcin, S. (2010). The nutritive value of modified dried vinasse (ProMass) and its effects on growth performance, carcass characteristics and some blood biochemical parameters in steers. *Revue de Médecine Vétérinaire, 161*, 245-252.

Zali, A., Eftekhari, M., Fatehi, F., & Ganjkhanlou, M. (2017). Effect of vinasse (condensed molasses solubles) on performance and meat chemical composition of Holstein male calves. *Italian Journal of Animal Science, 16*(3), 515-520.