Nigerian Journal of Agriculture, Food and Environment. 11(4):71-74

Published December, 2015

ORGANIC ACIDS AND ANTI-NUTRIENTS OF GUINEA GRASS (*Panicum maximum*) ENSILED WITH BREWERS' SPENT GRAINS

Ifut**, *O. J., Ekanem, N. J., Urang, S. H. and Inyang, U. A.

ABSTRACT

*Department of Animal Science, University of Uyo. * Corresponding author: dr.okonifut@gmail.com +2348037445486.*

This research sought to determine the organic acids and anti-nutrients of Panicum maximum (PM) ensiled with or without Brewers' spent grains (BSG). PM and BSG were mixed for silage preparation as follows: D1 = 100 % PM, D2 = 90 % PM + 10 % BSG, D3 = 80 % PM + 20 % BSG, and D4 = 70 % PM + 30 % BSG. The organic acid contents were 2.16 – 3.60, 1.20 – 2.40, 0.12 – 0.13 and 0.07 % for lactic, acetic, butyric and propionic acids respectively; they were within acceptable range for well-made silage. There were significant differences (P<0.05) in the lactic and acetic acid contents with increased level of BSG inclusion. The values obtained for ammonium nitrogen, phytate, saponin and tannin were 1.49 – 2.80, 0.02, 1.08 – 1.17 and 0.05 – 0.09 % respectively. Only the ammonium nitrogen content differed significantly (P<0.05), increasing with increased BSG addition. Although all the silages had good fermentation and chemical composition, D4 with the highest lactic acid, acetic acid and ammonium nitrogen contents would be better supplementary feed for ruminants.

Keywords: organic acids, anti-nutrients, Panicum maximum, brewer's spent grains, silage.

INTRODUCTION

The use of agricultural and agro-industrial by-products that have little or no food value to man are promising to alleviate nutrient deficiencies during the off-season (Babayemi *et al.*, 2010). Guinea grass *(Panicum maximum,* PM*)* is a major pan-tropical grass used throughout the tropics for pasture, silage and hay. It is a fast growing leafy grass, palatable and nutritious to livestock. However, it is generally recommended to supplement it with sources of protein in order to meet nutritional requirements or improve animal performance. Brewers' spent grains (BSG), the by-product of the grains fermented during beer making process, have been used as feed stuffs to both ruminants and monogastric animals (Westendorf and Wohit, 2002; Ekanem, 2012). BSG is considered to be a good source of undegradable protein and water soluble vitamins which stimulate appetite and feed intake (Tolkamp, 1988). Ensiling, chemical addition and drying into hay are some of the methods that may be used in preserving these feed stuffs. The quality of the ensiled product depends on the fermentation products, the types of acids and the amount of ammonia nitrogen present. Ensiling has been reported as one of the best methods in reducing the anti-nutritional factors such as phytate, saponin, tannin and ammonium nitrogen present in the material (Akinmutimi, 2004). The objectives of the study, therefore, were to determine the organic acid contents of PM ensiled with varying quantities of BSG and the effect of ensiling on the anti-nutrients composition of the silages.

MATERIALS AND METHODS

This research work was carried out at the Goatry Unit of the Department of Animal Science, Faculty of Agriculture, University of Uyo, Uyo, Akwa Ibom State between April and June, 2012. Uyo is located within the tropical rainforest zone which characterizes the South South agro-ecological zone of Nigeria. Uyo is located between latitudes 4°59′ and 5°04′ and longitudes 7°53′ and 8°00′ E. The *Panicum maximum* was collected from the University of Uyo Teaching and Research Farms, Use-Offot, which is on an elevation of about 60.96 m above sea level. The harvested grass was wilted for 24 h before chopped into uniform length of 2 – 3 cm and mixed with BSG which was sourced from the Akwa Ibom State Champion Breweries PLC. The ingredients composition of the treatments is shown in Table 1.

Table 1: Ingredients composition (%) of the experimental treatments

Ingredients	D1	D2	D3	D4
Panicum maximum (PM)	100	90	80	70
Brewer's spent grain (BSG)	0	10	20	30

The grass (PM) and brewer's spent grain (BSG) were mixed for silage in a 20-liter capacity plastic silo, with three replicates per treatment. Each silo was lined with polythene sheets, quickly filled, compressed to eliminate trapped air, and rapidly sealed to prevent re-entry of air. Sand bags were placed on top of each compressed and

polythene sheet-covered mass to further expel air pockets. The lids of the silos were then put in place to prevent further air entry. The ensilage was for 28 d.
Fresh samples of the feed materials were oven-dried at 60 °C for 48 h and subsequently hammer milled to pass through 2 mm sieve. Dried and ground samples of the silage were used for chemical analysis. The proximate composition of the silages was determined according to methods of A.O.A.C. (1990). Acid detergent fibre, neutral detergent fibre and acid detergent lignin were determined according to the method of van Socst *et al.,* (1991). Lactic acid and acetic acid were determined by the method of A.O.A.C. (1990). The absorbance or optical density of sample extract and propionic acid working standards were read at a wavelength of 515nm on a Spectronic 2ID spectrophotometer (Kayol and Borilek, 1995). The absorbance of sample extract and butyric acid working standards were read at wavelength of 570 nm on a Spectronic 2ID spectrophotometer (AOAC, 1990; Kayol and Borilek, 1995). Tannin was determined by Folin-Dennis Spectrophotometric Method (Pearson, 1976). The saponin content of the sample was determined by the Double Solvent Extraction Gravimetric Method (Harborne, 1980). Phytate was determined according to the method of McCance and Widdowson (1953), while Ammonia Nitrogen (NH_3N) was determined according to Nessler's Colorimeter Method (A.O.A.C., 1990). Data collected were statistically analyzed according to the Statistical Analysis System package (SAS, 1999). Significant means were separated using Studentized t-test and Duncan's Multiple Range Test of the same package.

RESULTS AND DISCUSSION

The chemical composition of the PM and BSG used for the ensiling is shown in Table 2. There were significant differences among the chemical composition parameters of the two feed stuffs used in ensiling ($P<0.05$), except for CF and NDF. The DM content in BSG (43.3 %) compared favourably with 42 % reported by O'Briens (2007), while that of PM (38.3 %) was within the range (35 – 45 %) obtained by Muller (2003). The CP obtained from PM and BSG were above the critical CP level of 8 % (Nortón, 1994) acceptable for ruminant performance. The ash content obtained (6.0 % for PM and 2.9 % for BSG) were lower than the 12 % reported by Omole *et al.* (2011). The values for the fibre fractions were within acceptable range reported by Wright (1999) and Fajemilehin *et al.* (2008). The result of the silage organic acids is presented in Table 3. The highest amount of lactic acid (3.6 %) was recorded in D4 while the least (2.16 %) was observed in D1 and D2 respectively. This showed the presence of lactic acid bacteria (LAB). They caused a drop in silage pH. There were significant differences ($P<0.05$) in the lactic acid content of the silage with increased inclusion of BSG. Since lactic acid retained energy in the form of fermented carbohydrates, it showed the presence of energy in the silage. This energy is needed by the animals for daily metabolic activities (van Soest, 1982). Acetic acid content of 2.40 % was recorded for D4 followed by D3 (2.2 %) and D2 (1.6 %) and the least (1.2 %) was D2. Acetic acid increased in the diets with increased inclusion of BSG. D2 (90 % PM + 10 % BSG) differed significantly ($p<0.05$) from D1 (100 % PM). There was no significant difference ($p>0.05$) between D3 and D4. However, D4 was numerically higher than D3. The range of values obtained in this study (1.55 – 2.40 %) is within the range of 1 - 3 % reported by Piltz and Kaiser (2004) for good quality silage. The propionic acid obtained for all the silages was 0.07 %. This is in agreement with Piltz and Kaiser (2004), who suggested that for good silage, the value of propionic acid should range between less than 0.1 to 0.3 percent. The values of butyric acid for D1, D2, D3 and D4 were 0.12, 0.13, 0.13 and 0.13 % respectively. The range of values obtained were in agreement with Piltz and Kaiser (2004) who stated that good quality silage should have butyric acid amount less than 0.5 %.

Table 2: Chemical Composition (%) of PM and BSG Used for the Ensiling (As fed basis).

Parameters (%)	PM	SE	BSG	SE
Dry matter (DM)	38.33[b]	1.17	43.29[a]	0.69
Crude protein (CP)	12.60[b]	0.69	23.80[a]	0.64
Crude fibre (CF)	4.04[a]	0.58	2.00[a]	0.29
Ether extract (CF)	2.60[b]	0.69	9.64[a]	0.69
Ash	6.00[a]	0.58	2.92[b]	0.23
Nitrogen free extract (NFE)	74.76[a]	1.27	61.64[b]	0.69
Acid detergent fibre (ADF)	49.88[a]	0.55	43.14[b]	0.02
Neutral detergent fibre (NDF)	64.86[a]	0.20	65.30[a]	0.42
Acid detergent lignin (NDL)	24.74[a]	0.22	19.75[b]	0.06

[a,b] Means on the same row with different superscripts differ significantly ($P<0.05$),
SE = Standard Error.

The result of the silage organic acids is presented in Table 3. The highest amount of lactic acid (3.6 %) was recorded in D4 while the least (2.16 %) was observed in D1 and D2 respectively. This showed the presence of lactic acid bacteria (LAB). They caused a drop in silage pH. There were significant differences ($P<0.05$) in the

lactic acid content of the silage with increased inclusion of BSG. Since lactic acid retained energy in the form of fermented carbohydrates, it showed the presence of energy in the silage. This energy is needed by the animals for daily metabolic activities (van Soest, 1982). Acetic acid content of 2.40 % was recorded for D4 followed by D3 (2.2 %) and D2 (1.6 %) and the least (1.2 %) was D2. Acetic acid increased in the diets with increased inclusion of BSG. D2 (90 % PM + 10 % BSG) differed significantly (P<0.05) from D1 (100 % PM). There was no significant difference (P>0.05) between D3 and D4. However, D4 was numerically higher than D3. The range of values obtained in this study (1.55 – 2.40 %) is within the range of 1 - 3 % reported by Piltz and Kaiser (2004) for good quality silage. The propionic acid obtained for all the silages was 0.07 %. This is in agreement with Piltz and Kaiser (2004), who suggested that for good silage, the value of propionic acid should range between less than 0.1 to 0.3 percent. The values of butyric acid for D1, D2, D3 and D4 were 0.12, 0.13, 0.13 and 0.13 % respectively. The range of values obtained were in agreement with Piltz and Kaiser (2004) who stated that good quality silage should have butyric acid amount less than 0.5 %.

Table 3: Organic acid contents (%) of ensiled *Panicum maximum* with brewer's spent grains at day 28.

Parameters	D1	D2	D3	D4	SEM
Lactic acid	2.16^{c}	2.16^{c}	3.24^{b}	3.60^{a}	0.04
Acetic acid	1.55^{b}	1.20^{c}	2.16^{a}	2.40^{a}	0.08
Propionic acid	0.07^{a}	0.07^{a}	0.07^{a}	0.07^{a}	0.00
Butyric acid	0.12^{b}	0.13^{a}	0.13^{a}	0.13^{a}	0.00

^{a,b,c,} means on the same row with different superscripts differ significantly (P<0.05).
Diet1= 100 % PM, Diet 2= 90 % PM + 10 % BSG, Diet 3 = 80 % PM + 20 % BSG, Diet 4 = 70 % PM + 30 % BSG.
SEM = Standard Error of Mean

Anti-nutrient contents of the diets

The result of the silage anti-nutrients is presented in Table 4. The highest amount of ammonium nitrogen (2.80 %) was observed in diet 4. The least value (1.49 %) was observed in diet 2. The trend showed an increase in NH_4N with increased brewer's spent grains inclusion. Diet 4 differed significantly (P<0.05) from diets 1, 2 and 3. The values fell within the range of excellent silage (less than 5 %) suggested by NSW (2008). Phytic acid percentage of 0.02 was observed for all the silages. The brewer's spent grains did not influence the phytic content of the ensiled materials. The result was within the range (0.02 – 0.03 %) for a well-made silage reported by Reddy (2001), but lower than the toxic level (590 – 750 mg/100 g DM) reported by Ene-Obong (2006). Saponin had its highest value (1.17 %) in silage 4 and the least (1.08 %) in silage 3. There was no significant differences (P>0.05) among the diets irrespective of their values numerically and the percentage inclusion of BSG. The percentage saponin obtained in this study was within the range of 1.00 – 2.99 % for ruminants, but below the toxic level of 3 % reported by Givens (2000). The highest percentage of tannin (0.09 %) was observed in silage 1 and decreased numerically to silage 4 (0.05 %), decreasing with increased level of BSG inclusion. There were no significant differences (P>0.05) among the diets. When compared to the moderate levels (<4 %) reported by Givez- Chavez (1996) and to the toxic level of 20 % for ruminants as reported by Harborne (1996), the composition of tannin in this study was below toxic level and hence safe for the livestock

Table 4: Anti-nutrients composition (%) of ensiled PM with BSG at day 28

Parameters (%)	D1	D2	D3	D4	SEM
NH_4N	1.96^{b}	1.49^{c}	2.05^{b}	2.80^{a}	0.09
Phytate	0.02	0.02	0.02	0.02	0.00
Saponin	1.16	1.15	1.08	1.17	0.04
Tannin	0.09	0.08	0.07	0.05	0.02

^{a,b,c,} Means on the same row with different superscripts differ significantly (P<0.05).
Diet1= 100 % PM, Diet 2 = 90 % PM + 10 % BSG, Diet 3 = 80 % PM + 20 % BSG, Diet 4 = 70 % PM + 30 % BSG.
SEM = Standard Error of Mean. NH_4N = Ammonia nitrogen.

CONCLUSION

The organic acid contents obtained in all the silages were within the range of acceptable silage. Silage 4 with the highest value of lactate and acetate will make a better feed for ruminants. The ensiling process reduced the amounts of anti-nutritional factors (ammonium nitrogen, phytate, saponin and tannin) of the feedstuffs thus making nutrients available.

REFERENCES

A.O.A.C. 1990. *Official Methods of Analysis, (15th edition). Published by the Association of Official Analytical Chemists,* Washington DC, USA.

Akinmutimi, A. H. 2004. Evaluation of Sword beans (*Canavalia gladiata)* as an alternative feed resource for broiler chickens. Ph.D. Thesis, Michael Okpara University of Agriculture, Umudike.

Babayemi, O. J., Ifut, O. J., Inyang, U.A. and Isaac, L. J. 2010. Quality and Chemical Composition of cassava waste ensiled with *Albizia sama pods. Agricultural Journal,* 5(3): 225 – 228.

Ekanem, N. J. 2012. Silage characteristics and Performance of Red Sokoto goats fed cassava peels ensiled with brewer's dried grains and poultry Litter M.Sc. Thesis Submitted to the Department of animal Science. Faculty of Agriculture University of Ibadan September 2012.

Ene-Obong, H. N. 2006. Content of anti-nutrients and invitro protein digestibility of the African yam bean, pigeon and cowpea. *Plans Food for Human Nutrition,* 62(2):106 – 120.

Fajemilehin, S. O. K., Babayemi, O. J., Fagbuaro, S. S. 2008. Effects of anhydrous magnessium sulphate fertilizer and cutting frequency on yeild and chemical composition of *Panicum maximum. Afri. J. of Biotech.* Vol. 7(7): 907 – 911.

Givens, D. I. 2000. *Forage Evaluation in Ruminant Nutrition.*

Givez-Chavez, B. I. 1996. Condensed tannins in tropical forages. Cornell University, Ithaca, NY, USA.

Harborne, J., Alan, R. L. 1996. An alternative and faster isolation method. Pp. 67 – 76, New York.

Kayol, L. T. and Borilex, G. O. D. 1995. Spectrophotometric Determinations of Volatile fatty acids. *Anal. Chem.* 34: 112 - 117.

McCance and Widdowson, 1953. *Food Analysis using Spectrophotometer.*

Muller, J. 2003. Silage Insights – Pre – wilting and how to estimate dry matter content of grass silages. DLG - Information.

Norton, B. W. 1994. Tree legumes as dietary supplments for ruminants. In: Gutteridge, R. C., Shelton, H.M. (editors). Forage tree legumes in Tropical Agriculture, CAB International. pp. 192 – 201.

NSW. 2008. Feed Quality Service-Interpreting the Fed Analysis Report. A Work by NSW Department of Primary Industries.

O'Briens, C. 2007. Grains of Possibility: Ways to use spent brewing grains. *American Brewer Magazine.* Spring 2007 Edition.

Omole, A. J., Ogungbesan, A. M., Fayenuwo J. A., Popoola Y. A. 2011. Comparative utilization of *Centrosema pubescens* or Mucuna purensis as a substitute for pawpaw leaf by growing snails. *Lives. Res. For Rur. Dev.* Volume 23, Article #63. http://www.lrrd.org/lrrd234/3/omol24063.htm.

Pearson, D. A. 1976. *Chemical Analysis of Foods.* 7th Ed. Churchill Livingstone. Edinburgh.

Piltz, J. W. and Kaiser, A. G. 2004. Principles of silages preservation. In: A.G. Kaiser, J. W. Piltz, H. M. Burns and N. W. Griffiths (eds). Top Fodder Successful silage. Pp. 25 -26. Dairy Australia and New South Wales Department of Primary Industries Orange, NSW.

Reddy, N. R. 2001. Food Phytates. 1st Ed. pp. 30 – 32, 118. *Journal of Agric. and Food Sci.,* 49 (5): 2657 – 2662. CRC Press.

Statistical Analysis System (SAS) 1996. SAS/STAT User's guide, SAS Institute Inc. Cary, North Carolina, USA.

Tolkamp, B. 1988. Feed intake regulation and small ruminants: Goat production in the humid tropics (Eds. O. B. Smith and H. G. Bosman) *Pudoc.* Wageningen. Pp. 67 – 75.

Van Soest, P. J. 1982. Nutritional Ecology of the Ruminants, ruminant metabolism, nutritional strategies, the cellulolytic fermentation and the chemistry of forages and plant fibres. Cornell University, United States of America.

Van Soest, P. J; Robertson, J. P. and Lewis, B. A. 1991. Methods for determing dietary fibre nentural detergent fibre and non-starch. Polysaceha rides in relation to animal mutation J. Daily Sci J4: 3583-3597.

Westendorf, M. L. and Wohit, J. E. 2002. Brewing by-products their uses as animal feeds . *Vet. Clin. Food Animal Practical* 18: 233 - 252.

Wright, F. 1999. Grass Silage Analysis Reports and Explanation. Quality Products for Quality Production. GP Feeds Ltd.