IN VITRO GAS PRODUCTION AND DIGESTIBILITY OF CASSAVA-BASED CONCENTRATE INCUBATED WITH YEAST (*Saccharomyces cerevisiae*)

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Abstract
The study evaluated the effect of varying levels of *Saccharomyces cerevisiae* on the *in vitro* digestibility of neutral detergent fibre, dry matter, post incubation parameters and fermentation kinetics of cassava based diet. The treatments are inclusion of yeast at 0 g/kg, 1.6 g/kg, 3.3 g/kg, and 5 g/kg of concentrate diet. The experiment was set up in a completely randomized design. Data obtained were analysed using one way analysis of variance. The result obtained shows that gas production, organic matter digestibility (OMD) and short chain fatty acid (SCFA) were significantly ($p<0.05$) influenced across the treatment as the level of Yeast. Organic matter digestibility was highest at 3.3 g/kg SC inclusion level and lowest at 1.6 g/kg/kg SC inclusion level. Short chain fatty acid has highest value at control group and 3.3 g/kg SC inclusion level and lowest at 1.6 g/kg/kg SC inclusion level. The neutral detergent fibre digestibility (NDFD) increases progressively across the treatment (62.68-67.38) at varying level of inclusion. Inclusion of Yeast at 0.5-5 g/kg significantly increased NDFD and DMD. The result obtained shows that supplementation of Yeast gave the best in improving the digestibility of cassava based diet at the level of 1.6 g/kg/kg of dry matter ration.
Keywords: Yeast, *in vitro*, gas production, digestibility.

Introduction
The productive performance of ruminants is a function of the nutritional value of the diet they consume. Ruminants have a unique ability to convert feedstuff of low nutritional value or unfit for human consumption into useful end products that are utilized for productivity and growth. For optimal utilization of feed, it would be desirable to be able to quantify and express, in feeding standard, the needs for rumen micro-organisms and for the host animal separately (Czerkawski, 1986). Aside from the natural capacities of the rumen microbes to degrade fibrous feedstuffs and express their nutrient both to the host and the microbes themselves, several organic and inorganic compounds have been used to engineer the activities of the microbes and improve rumen function generally (Dawson and Newman, 1987). Digestive processes in the rumen can be manipulated through the addition of direct-fed microbial which enhances feed digestion, to improve the performance of animals and to boost the health status of animal (Robinson and Erasmus, 2009). Yeast products for ruminants based on *Saccharomyces cerevisiae* increase the number of cellulolytic bacteria (Wallace and Newbold, 1993; Alzahal *et al*., 2014), and are associated with a higher rumen pH promoted by the yeast, which favours the growth of fibrolytic bacteria *(Fibrobacter and Ruminococcus)* and lactate-utilising bacteria (*Megasphaera* and *Selenomonas*; Pinloche *et al*., 2013). The objective of this study was to access the effects of varying levels of yeast on *in vitro* gas production and digestibility of cassava-based concentrate.

Materials and Methods
The experiment was carried out at Animal Nutrition Department Laboratory, Federal University of Agriculture Abeokuta, Ogun state, Nigeria. The experimental design was a completely randomized design (CRD) with four levels of yeast (0 g/kg, 1.6 g/kg, 3.3 g/kg, and 5 g/kg) added to the concentrate feed (cassava peel (30%), dried brewers grain (30%), rice bran (30%), bone meal (5%) and salt (5%)). Rumen fluid was collected from three (3) cattle from Cattle Production Venture, FUNAAB by using suction tube prior to morning feeding. The method of collection was as described by Babayemi *et al.* (2006). The ground

samples were stored in plastic bags until used for experimental procedures. Rumen liquor was collected from cattle into warm insulated flasks and used as the source of inoculums. The donor animal was maintained daily on ad libitum *Panicum maximum*. The *in vitro* gas production procedure by Menke and Steingass (1988) was employed for the experiment. A sensitive scale was used to measure 200mg of substrate, replicated five times and were placed into 50ml calibrated plastic syringes fitted with plungers. The rumen fluid (inoculum) was filtered through layers of cheesecloth and mixed with buffer at a ratio of 1:2 (v/v), and resazurin dye was added to sustain the activities of microbes. Fifteen millilitres of the buffered inoculums was then added to each syringe containing the milled samples and the gas released was read directly on the graduated syringe. The syringes were placed in an incubator kept at 40°C along with blank syringes containing only 15ml of the buffered inoculums. The syringes were shaken at regular intervals to simulate rumen motility. Gas production was recorded for 48 hours of incubation at 3-hour intervals. Organic matter digestibility (OMD) was estimated as OMD = 14.88 + 0.889 GV + 0.45 CP + 0.651 ash (Menke and Steingass, 1988).
Short-chain fatty acids (SCFA) was estimated as SCFA = 0.0239 GV − 0.0601 (Getachew et al., 2000). Metabolizable energy (ME) was calculated as ME = 2.20 + 0.136 GV + 0.057 CP + 0.029 CP^2 (Menke and Steingass, 1988). Where, GV was the net gas production at 24 hrs and CP was crude protein. Gas volume at 24 hrs was expressed as ml/0.2 g DM, CP and ash as g/kg DM, ME as MJ/kg DM, OMD as % and SCFA as µmol/g DM. Data obtained from this experiment were analysed using one-way analysis of variance option of the SPSS (IBM SPSS Statistics 20) software. Treatment means were statistically compared using Duncan's Multiple Range Test to identify differences between means and significant differences were declared if $p<0.05$.

Results and Discussion

Table 1 shows the gas production values of diets incubated with varying levels of yeast. Gas values showed that higher ($p<0.05$) values were recorded with inclusion of yeast (9.38 ml vs 6.89 ml at 6 hours of incubation for 5 g/kg and 0 g/kg inclusion respectively). However, with longer duration of incubation (24 hours), the highest ($p<0.05$) gas production was recorded with 3.3 g/kg (19.38 ml) while lower levels were recorded with 1.6 g/kg (15.88 ml) and 5 g/kg (17.38 ml).

Table 1:Gas production values of diet incubated with varying levels of yeast (*Saccharomyces cerevisiae*)

	Levels of yeast inclusion					
Incubation time	0 g/kg	1.6 g/kg	3.3 g/kg	5 g/kg	SEM	P value
3	3.50	3.13	3.50	3.63	0.083	0.18
6	6.89^{ab}	7.31^{b}	8.00^{ab}	9.38^{a}	0.168	0.05
12	13.44^{ab}	12.13^{b}	14.25^{a}	14.50^{ab}	0.267	0.04
24	19.00^{a}	15.88^{b}	19.38^{a}	17.38^{ab}	0.444	0.01

^{ab} Means on the same row with different superscripts are significantly different ($P<0.05$)

Table 2 shows the *in vitro* digestibility and post-incubation parameters of cassava based diet incubated with varying level of yeast. The OMD was particularly highest at yeast inclusion level of 3.3 g/kg but the value were similar at control group and 5 g/kg. The short chain fatty acid (SCFA) values were low at varying level of inclusion of yeast. This was due to lower gas production which was evident in the first 24hour of incubation. The level of SCFA is an indicator of energy availability to the animal. OMD and SCFA were significantly ($p<0.05$) influenced as the yeast inclusion level increases across the treatment. The OMD were

similar across the treatment. Also the SCFA were similar at yeast inclusion level of 0 g/kg, 3.3 g/kg, and 5 g/kg but different at the 1.6 g/kg. The value for the SCFA ranged from 0.25-0.36. Gas production was directly proportional to SCFA (Beuvink and Spoelstra, 1992). Higher SCFA value at the control group and 3.3 g/kg yeast inclusion level indicates potential of the feed to make energy available to the ruminants.

Table 2: *In vitro* digestibility and post-incubation parameters of cassava based diet incubated with varying level of yeast

	Levels of yeast inclusion					
Parameters	0 g/kg Yeast	1.6 g/kg Yeast	3.3 g/kg Yeast	5 g/kg Yeast	SEM	P value
GP	19.00[a]	15.88[b]	19.38[a]	17.38 [ab]	0.444	0.01
NDFD	62.68[b]	67.38 [a]	67.16[a]	66.37[a]	0.561	0.01
DMD	16.88[b]	20.92 [a]	21.13 [a]	18.20[ab]	0.689	0.02
OMD	38.36 [a]	34.34 [a]	38.56 [a]	38.12[a]	0.666	0.02
SCFA	0.36 [a]	0.25[b]	0.36 [a]	0.35 [a]	0.179	0.02
ME	10.66	10.02	10.66	10.60	0.127	0.21

[ab] Means on the same row with different superscripts are significantly different (P<0.05)
NDFD-Neutral Detergent Fibre, DMD-Dry Matter Digestibility, GP-Gas Production, OMD-Organic Matter Digestibility, SCFA-Short Chain Fatty Acid, ME-Metabolisable Energy

The chemical composition of food depicts its nutritive value, and the digestibility is a measure of the nutritional usefulness of feed or availability of the nutrient component to the fed species.
Inclusion of yeast at 1.6 – 5 g/kg g significantly increased NDFD and DMD. Plata *et al.* (1994) found also that NDF digestibility increased with yeast addition (48.6 vs. 60.45). Some of the benefits associated with yeast include increased DM and the neutral detergent fibre digestibility (NDFD) increases progressively across the treatment (62.68 – 67.38) at varying level of inclusion. Inclusion of Yeast at 1.6 – 5 g/kg g significantly increased NDFD and DMD, NDF digestion; in addition, the yeast increases NDF digestibility and the number of protozoa present in the rumen and increased the microbial protein flow from the rumen in lactating dairy cattle and in buffalo bulls. Increased digestibility of NDF with supplementation of yeast caused by increased population especially rumen cellulolytic microbes (Callaway and Martin, 1997).
The result obtained shows that supplementation of yeast at the inclusion level of 1.6 g/kg gives the best result in improving the digestibility of cassava based diet. The value of the dry matter digestibility at varying level of yeast, ordered from the lowest to the highest was 0, 1.5, 0.5 and 5 g/kg g.
The values of organic matter digestibility at varying level of SC ranged from 34.34 to 38.56% and these values are greater than the ones of dry matter digestibility. The highest average of the organic matter digestibility values (38.56%) was obtained at inclusion level of 3.3 g/kg of SC. According to Firsoni, *et al.* (2008) the complete feed of organic matter digestibility ranged from 48.32 to 53.75%. Therefore, in general, organic matter digestibility was under normal. The digestibility of organic matter looks the best among all the treatment at 3.3 g/kg SC inclusion level. This is because the mixture of concentrate with yeast achieves the required balance of nutrients which is needed by rumen microbes so that they can grow optimally and easily degrade organic matter.

Conclusion and Recommendations

From the results of this study, it can be concluded that the addition of yeast improves the neutral detergent fibre digestibility of the cassava based diet. Dry matter digestibility was also improved, for this will achieves the required balance of nutrients which is needed by rumen microbes so that they can grow optimally and easily degrade organic matter. Higher SCFA value obtained indicates the potential of the feed to make energy available to the ruminant animals. Yeast supplementation at varying levels of inclusion improves

the organic matter digestibility. Further studies are therefore recommended to investigate higher levels of yeast inclusion and the interaction between yeast and protozoa with its bearing on methane production

References

Alzahal, O. Dionissopoulos, L., Laarman, A. H., Walker, N. and McBride, B. W., 2014. Active dry Saccharomyces cerevisiae can alleviate the effect of subacute ruminal acidosis in lactating dairy cows. *J. Dairy Sci.* 97, 7751-7763.

Babayemi, O. J., 2006. Antinutritional factors, nutritive value and in vitro gas production of foliage and fruit of *Enterolobium cyclocarpum*. *World J. Zool*., 1(2): 113-117.

Beuvink, J. M. and Spoelstra, S. F., 1992. Interaction between treatment, fermentation end-products buffering systems and gas production upon fermentation of different carbohydrates by mixed-microorganism *in vitro*. *Appl. Microbial Biotechnol.*37:505;509.

Callaway, E. S and S.A. Martin 1997. Effects of a *Sccharomyces cerevisiae* culture on ruminal bacteria that utilize lactates and digest cellulose. *J. Dairy Sci.* 80:2035-2044.

Czerkawski J. W., 1986. An introduction to Rumen Studies. Oxford, Pergamon Press. Pp 122-132.

Dawson, K. A. and Newman, K. E., 1987. Effects of yeast cultures supplement on the growth and activities of rumen bacteria in continuous cultures. *J. Anim. Sci.*

Plata, P. F., Mendoza, M. G. D., Gama, J. R. B., and Gonzalez, M. S., 1994. Effect of a yeast culture (Saccharomyces cerevisiae) on neutral detergent fibre digestion in steers fed oat straw based diets. *Animal Feed Science and Technology*

Pinloche, E., McEwan, N., Marden, J. P., Bayourthe, C., Auclair, E. and Newbold, C. J., 2013. The effects of a probiotics yeast on the bacterial diversity and population structure in the rumen of cattle. PLOS One 8:e67824.

Robinson, P.H and Erasmus, L.J., 2009.Effect of analyzable diet components on response of lactating dairy cows to *Saccharomyces cerevisiae* based yeast product: A systematic review of the literature. *Anim. Feed Sci. Technol*. 149:185-198.

Menke, K.H. and Steingass, H., 1988. Estimation of the energetic feed value obtained from chemical analysis and in vitro gas production using rumen fluid. Anim. Res. Dev. 28: 7–55.

IBM SPSS 2011. Statistics Version 20. IBM Corporation.

Wallace, R. J. and Newbold, C. J., 1993. Rumen fermentation and its manipulation: the development of yeast cultures as feed additives. In: Biotechnology in the Feed Industry. Ed: Lyous, T.P., All tech Technical Publications, Nicholasville, Kentucky. pp. 173-192.