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Apparent digestibility of solid state fermentation of cotton waste with fungus (*Pleurotus sajor caju*) using West African dwarf goats

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ABSTRACT: The effects of consumption of fungus treated cotton waste by growing goats on feed intake and digestibility was determined. Nine West African dwarf goats (7.5 ± 0.65 kg average trial BW) in a 3 x 3 Latin square with 21d periods, consumed cotton seed cake diet (diet A), cotton seed meal (diet B) and fungus treated cotton waste (diet C) *ad libitum*. Cotton seed cake, cotton seed meal and treated cotton waste diets were 2.66, 2.24 and 2.91% N and 30.62, 28.64 and 26.05% NDF, respectively. ADF was 19.87, 17.59 and 16.00% in diets A, B and C respectively. Total organic matter intake (OM) was 268.93, 265.10 and 287.43 gld. Dry matter and crude protein digestibilities increased more for diet C ($P < 0.05$) than the mean for diets A and B. Crude fibre digestibility ranked ($P < 0.05$) B (70.41%) > A (65.7%) > C (58.20%) and ADF digestibility was 50.25, 60.66 and 74.20% (± 6.75 SE). NDF was greatest ($P < 0.05$) for diet C, greater ($P < 0.05$) for diet B than for diet A (67.70, 58.84 and 42.20% respectively).

In conclusion, fungus treatment of cotton waste for growing goats may improve digestibility of nutrients as a result of higher digestibilities of dry matter, crude protein, acid detergent fibre (ADF) neutral detergent fibre (NDF) and hemicellulose. The possibility that fungi may enhance the crude protein content and reduced the fibre fractions further confirmed the propriety of this study.

Key Words: Food intake; Apparent digestibility; Fungus treated cotton waste; *Pleurotus sajor caju*.

Introduction

By the year 2020, the global population is expected to increase by more than 40%, possibly exceeding the billion mark. Feeding these additional population, with a limited supply of suitable farmland and livestock, poses a problem of immense proportions in the developing countries, not the industrial countries, where 800 million people suffer from malnutrition today (Jerven, 2000). The benefits of agricultural biotechnology are of particular importance to people living in developing nations in which crop and animal productivity could be increased thereby contributing to the alleviation of hunger and poverty which are inextricably linked (James, 1998). Biotechnology in the livestock production systems is available at three different points: feed before consumption; the digestion process within the animals and the metabolic processes in the animal (Jacqueline *et al*; 1996). However, one fairly widespread biotechnology process is

the addition of microbes and/or their products to animal feeds to improve their preservation and digestibility (Jacqueline *et al*; 1996). The treatment of lignocellulosic materials with fungi is one option that has received significant attention in recent years (Belewu and Okhawere, 1998; Belewu, 1999; Belewu and Banjo 1999; Belewu, 2000).

The most prominent and at present vital as lignin degrader is the mushroom. They exist as symbionts; saprophytes or parasites (Singer, 1954). The edible mushrooms which are saprophytic in nature can grow on a variety of cellulosic wastes (Chang, 1980; Garcha *et al*; 1984). The selective removal of lignin from lignocellulosic wastes by white fungi are well-documented (Kamra *et al* 1993, Belewu and Okhawere, 1998). However, little information exists in the literature on the feeding value of the spent substrate (i.e. the resulting materials after harvesting the matured fruit bodies) in ruminant nutrition. The objective of this study is, therefore, to evaluate the effect of spent substrate inclusion in the diet of ruminants on the digestibility of West African dwarf goats.

Materials and Methods

Source of Inoculum and Media Used

A culture of *Pleurotus sajor caju* was obtained from the culture collection of the Ruminant Nutrition and Biotechnology Laboratory, Department of Animal Production, University of Ilorin, Nigeria. The test organism was maintained on potato dextrose slant containing (g/l) potato infusion 200g, Bacto dextrose 20g and Bacto agar 15g.

Spawn preparation

The spawn was prepared according to the method of OEI (1996). Briefly, two litres of deionised water were added to 200g of sorghum grain and boiled in a large pot. It was later drained while additives like $CaCO_3$ (20g) and gypsum (80g) ($CaSO_4.2H_2O$) were added so as to stabilize the pH and have a positive effect on the structure of the substrate. The mixture was then scooped into dextrose bottles and the mouth was plugged with cotton wool. The filled bottles were later autoclaved at 121^oC for 1 hour on two successive days. The bottles were shaken when they were taken out of the autoclave to improve moisture uniformity and to keep the grain at the bottom from sticking together.

Inoculation

After cooling, the spawn was inoculated in an inoculating chamber with 10 x 10 mm^2 from the full-grown agar of the mother culture for each bottle. However, more pieces of the agar were added to speed up colonization.

Incubation

The bottles were incubated at 37^oC until the mycelium has grown all over the substrate. The bottles were shaken every three days to evenly distribute the mycelium and to prevent sticking of the substrate. The complete colonization took 2 weeks.

Storage

The spawn was kept in a refrigerator until needed for the inoculation of cotton waste.

Preparation of Cotton waste

The substrate used was cotton waste which was obtained from the Textile Mill in Lagos. The cotton waste was moistened with water and then transferred into polypropylene bags and sterilised in an autoclave at 121^oC for 1 hour. This pasteurised the cotton waste and killed any fast growing microbes.

Inoculating the cotton waste

The sterilised substrate was inoculated with a 5% (w/w) inoculum. Fifteen of such bags of ten kilograms each were obtained and incubated in a well-ventilated dark room. After 15 days incubation, the mycelia enveloped the substrate. The bags were occasionally watered to keep the substrate moist. About 30 days after inoculation, the immature fruit bodies developed and the matured fruit bodies were harvested as they mature. Total productive period was forty days from the date of inoculation until production ceased. The spent substrate was later oven-dried at 80^{o}C and used in the formulation of a diet for goats.

Preparation of the experimental diets and Animal Management

The spent substrate was included in diet C (19.80 %) while diets A and B contained cottonseed cake (19.80 %) and cottonseed meal (19.80 %). Other ingredients are as shown in Table 1.

Nine West African dwarf goats (7.5 ± 0.65 kg body wt.) were kept in wooden digestion crates to facilitate total faecal collection. A 3 x 3 Latin square designed was used with three goats per diet. Each diet was offered to three goats in each period *ad libitum* and each 7-day collection period was preceded by a 14 day adjustment period. Daily orts of the feed was taken and feed intake calculated while total faecal output was recorded and an aliquot (10%) taken, refrigerated during the collection period and frozen until analysed.

Table 1: Composition of the Experimental Diets

INGREDIENT (%)	Treatments		
	A	B	C
Cassava waste	50.10	50.10	50.10
Cotton Seed Cake	19.80	-	-
Cotton Seed meal	-	19.80	-
Treated Cotton Waste	-	-	19.80
Sorghum Dried Brewer's Grain	28.10	28.10	28.10
Salt	1.00	1.00	1.00
Vitamin-mineral premix	1.00	1.00	1.00
Total	100.00	100.00	100.00
Proximate Composition (%)			
Dry Matter	93.18	93.15	90.50
Crude Protein	16.60	14.02	18.20
Ether Extract	5.01	7.09	5.12
Neutral detergent fibre	30.62	28.64	26.05
Acid detergent fibre	19.87	17.59	16.00
Cellulose	12.05	13.45	11.10
Hemi-cellulose	10.75	11.05	10.05
Free Gossypal (%)	0.14	0.16	0.07
ME kcal	4.66	4.72	4.76

*Calculated from the proximate Composition.

Chemical analyses

The proximate composition of the feed and faeces were determined by standard A.O.A.C. (1990) procedures while the fibre fraction determined by the method of Goering and Vansoest (1970). The apparent digestion coefficients of the diets were calculated by the direct method as illustrated by Ranjhan (1980). Analysis of variance technique using a 3x 3 Latin square design model was used to evaluate data and comparisons among means were made by Duncan Multiple Range Test (Duncan, 1955).

Result and Discussion

The nitrogen concentration of diet C was considerably higher than that of diets A.& B. (Table1). Diet C was also higher in metabolizable energy (ME) than diet A but because of the fungus treatment, acid detergent fibre (ADF), natural detergent fibre (NDF), cellulose and crude fibre were lower for diet C. fungus treatment resulted in a product that was lower in gossypol and fibre fractions. The ether extract (EE) also increased slightly as a result of the fungus treatment.

Data on digestibility (Table 2) show that fungus treatment resulted in substantial increase in digestion of various organic component shown ($P < 0.05$). for example organic matter (DM) intake was 268.93, 265.10 and 287.43 gld which increased by 8% for diet C. Dry matter digestibility was highest for diet C than the mean of A and B. Treating the cotton waste with fungus resulted in improved crude protein and ether extract but crude fibre digestibility was depressed $P < 0.05$). The fibre fractions, NDF, ADF, lignin and hemicellulose were somewhat higher in the treatment diet c than in diets, A and B. Other researchers have reported increased crude protein and other fibre fractions digestibility when fungi treatment samples were fed (Belewu, 1999; Belewu and Banjo, 1999 Belewu, 2000).

Table 2: Apparent digestibility of the experimental diets.

NUTIENTS	Diets			
	A	B	C	± SEM
Dry matter (%)	67.20	60.74	75.45	10.68*
Crude Protein(%)	58.59	47.86	65.03	4.58 *
Crude Fibre (%)	65.27	70.41	58.20	3.30
Ether Extract (%)	60.66	68.66	72.36	2.57 NS
Acid Detergent Fibre (%)	50.25	60.66	74.20	6.75
Neutral Detergent Fibre (%)	42.20	58.84	67.70	4.69
Lignin (%)	34.99	41.66	48.21	5.36
Hemicellulose (%)	54.44	65.29	71.02	2.72

The higher digestion coefficients of dry matter and other organic matter could be due to the pre-digestion of the subtrate which may have enhanced better digestion and absorption. Jacqueline *et al*. (1996) also noted that treatment of substrate with microbes (fungi) may have enhanced the crude protein due to the addition of fungal protein while the pre-digestions by the fungi, promote better digestibility of the substrate due to the degradation of ligno-cellulose, particularly the recalcitrant lignin component. The present study indicates that cotton waste processed to reduce crude protein and ADF digestibilities of 65.03 and 74.20% had higher nutritional value than diets A and B. Thus, the method seems an appropriate one for promoting

the digestibility of low quality roughages. The chief advantage of the method is that a highly fibrous and poor quality substrate, such as cotton waste fed alone is not required and acceptable to animals, so treatment with mushroom which is a good lignin degrader is necessary to develop and upgrade such substrate. The true protein fraction of total nitrogen in diet C include protein microbial cell and fungal protein formed during treatment (Jacqueline, 1996). Therefore, sloe liberate of nitrogenous compounds from diet C may have occurred with high availability of fibrolytic microbes (Akbar *et al*; 1987) or minerals (Sanni, 1995) in the treated sample may have affected fibres digestion favourably.

Equations (1-3) describe the regression equation, derived from percentage digestibility of DM (Y) of the diets and their correlation's:

Diets

A	Y	=	814 + 67.15x	R	= -0.59
B	Y	=	650 + 60.23x	R	= -0.48
C	Y	=	702 + 77.10x	R	= 0.52

There is a negative correlation between dry matter digestibility and the percentage composition of the fibre. The dry matter digestibility of diets A and B agreed with the report of Ranjhan, (1980), Adegbola and Obioha (1982) that high fibre level of diet depressed digestibility of dry matter and other components of the feed. While the improvement in the dry matter and fibre fractions digestibility of diet C agreed with the generally accepted observation (Ranjhan, 1980; Adegbola and Obioha, 1982).

Conclusion

In this experiment, the major factor restricting organic matter digestibility when fibrous substrates were fed appeared to be the complex bond of three polymers, cellulose, hemicellulose and lignin. In this experiment fully or partial degradation of the complex bond by fungus improved the digestibility of such materials, characteristics of the substrate (cotton waste) appeared to alter effects of the composition and this compensated for higher digestibility of the treated waste than others.

Treatment of waste with fungus improved the feeding value by increasing the digestibilities of nitrogen and other fibre fractions.

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