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Short communication

Changes in the cellulose, sugar and crude protein contents of agro-industrial by-products fermented with *Aspergillus niger, Aspergillus flavus* and *Penicillium* sp.

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Changes in the protein, sugar and cellulose of three agro-industrial by-products after fermentation with *Aspergillus niger, Aspergillus flavus* and P*enicillium* sp. in solid state were determined. Cellulose was significantly reduced (P<0.05) in all the agro wastes by all the fungi after 14 days. Highest percentage reduction was achieved by *A. niger* in all the agro wastes; 36.51% for wheat offal (WO), 35.87% for brewers dried grain (BDG) and 35.80% for maize offal (MO). There was a corresponding build up of all the soluble sugars in the substrate up to 14 days. Beyond day 14, the sugar level began to fall and there was no more significant degradation of the cellulose. The crude protein of the BDG, MO and WO increased significantly (P<.0.05). After 14 days, the highest percentage increase in protein (41%) was obtained in WO inoculated with *A. niger.* Results of the study indicate the possibility of enhancing the feeding value of these by-products by a simple, inexpensive and easily adaptable technique.

Key words: Cellulose, sugar, protein, changes, fermentation, fungi.

INTRODUCTION

Agro-industrial by-products in Nigeria vary from primary processing of farm produce wastes to wastes form agro allied industries. Some of these wastes are left unutilised, often causing environmental pollution and hazard. Those that are utilized do not have their full potentials harnessed. Agro-industrial by products which can be of tremendous use in the livestock industry for feeding animals include brewers dried grain (BDG), palm kernel cake (PKC), maize offal (MO), wheat offal (WO) and cassava peels (CP).

As grain production remains insufficient to meet human and animal feeding, the alternative is to employ feed ingredients which do not have direct human value (Akinwumi, 1989). BDG, MO, and WO are by-products of sorghum, maize and wheat processing. They are of low protein and high crude fibre contents. These are two factors that limit their use in poultry and pig feeding, the sector of the livestock industry that constitutes the largest consumer of commercial feeds in Nigeria.

It is imperative to enhance the nutritive values of these by-products through the breakdown of their non-starch polysaccharides (NSPs). The use of microbial degrading enzymes from fungi to achieve this is possible and hightly desirable. The study reports the changes in the nutritive value of BDG, MO, and WO by *Aspergillus niger, Aspergillus flavus* and *Penicillium* sp.

MATERIALS AND METHODS

Fungi

The fungi used in the study were *A. niger, A. flavus* and *Penicillium* sp. Slants of the microbes were obtained from the culture bank of the Department of Microbidogy, University of Ibadan. A piece of mycelia of each of the fungi was then subcultured on potato dextrose agar (PDA) in petri dishes and incubated at 30^0C for 2 days.

Agro by-product as substrates

30 g of each of the agro by-products (BDG, MO and WO) were weighed into 250 ml conical flasks in triplicates and the moisture adjusted to 25%. The mouth of the flasks were clogged with cotton wool and then covered with aluminium foil. The flasks containing the substrates were autoclaved at 121^0C for 15 min. After

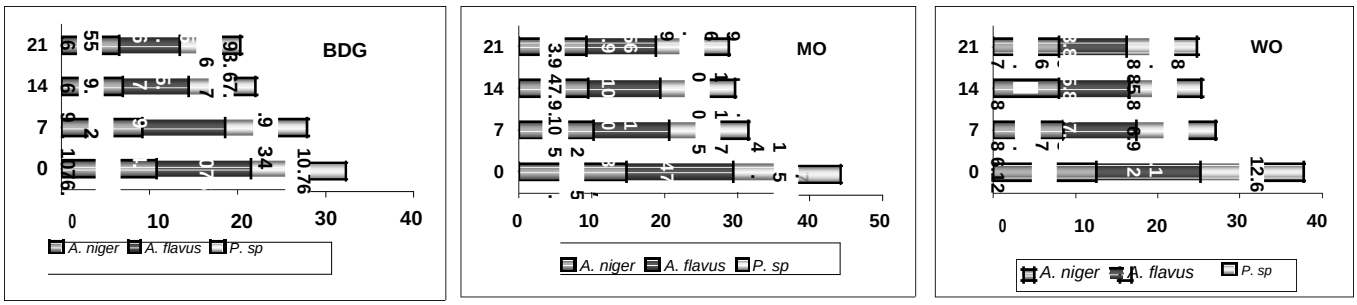

Figure 1. Changes in cellulose content in brewer's dried grain (BDG), maize offal (MO) and wheat offal (WO) after biodegradation with *A. niger, A. flavus* and *Penicillium* sp. (*P. sp*).

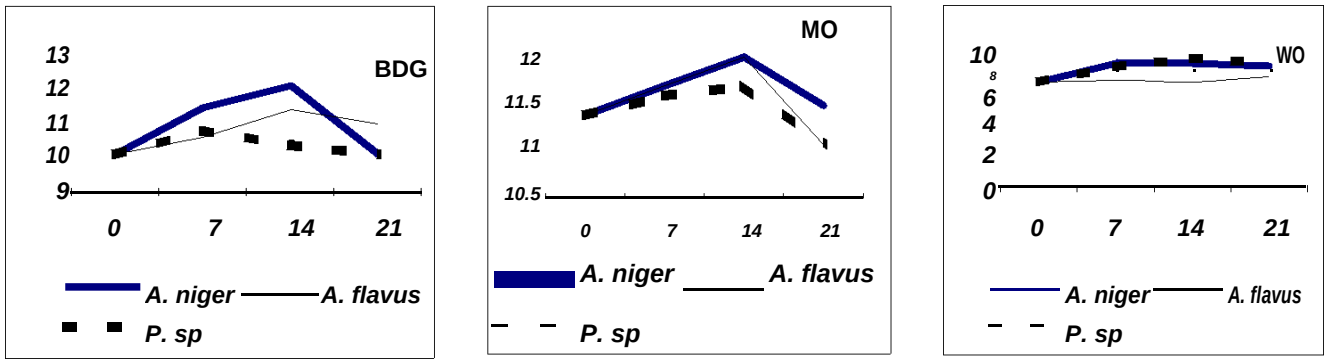

Figure 2. Changes in levels of soluble sugars in brewer's dried grain (BDG), maize offal (MO) and wheat offal (WO) biodegraded with *A. niger, A. flavus* and *Penicillium* sp. (*P. sp*).

autoclaving, a piece of mycelia from the subculture of the respective fungus was added to each flask. Three set of flasks each containing BDG, MO, and WO were aseptically inoculated in a lamina flow cabinet with each of the 3 fungi. Another set of 3 flasks containing the substrates was uninoculated. All the flasks were covered back and placed in an incubator set at 30^0C

Samples were withdrawn after 7, 14 and 21 days, the action of the microbes terminated by drying at 60^0C. The samples were thoroughly mixed, freeze-dried, milled and stored in sterilized bottles. Samples from control flasks were treated alike.

Analysis of samples

The samples were analysed for protein by the method of Lowry (1962), cellulose by the technique of Hendry and Grime (1973) and soluble sugar by the method of Deriaz (1961) and Dubous et al. (1952). The period at which minimum level of cellulose and maximum level of sugar were obtained in the media was taken as the optimum period (Iyayi and Losel, 2001). All data were subjected to analysis of variance (ANOVA) according to the procedure of Steels and Torrie (1960) and paired means separated by Duncan's multiple range test where there were significant differences.

RESULTS AND DISCUSSION

The results of changes in cellulose, soluble sugars and crude protein in BDG, MO and WO after 7, 14 and 12 days are presented in Figures 1, 2 and 3. Cellulose in BDG, MO and WO was significantly ($P<0.05$)) reduced by all the fungi after 14 days. Thereafter, reduction still occurred though not significant. *A. niger* consistently caused the highest reduction in cellulose in all the by-products followed by *A. flavus* and *Penicillium* sp. The highest perecentage reduction by *A. niger* was achieved in WB (36.51%) followed by BDG (35.87%) and MB (35.80%). The sugar production increased up to day 14 for all the by-products when inoculated with the fungi and thereafter fell. The ability of fungi to degrade cellulose has been reported elsewhere (Ofuya and Nwanjiuba, 1990; Iyayi and Losel, 2001). Earlier works of the first authors showed successful degradation of cassava peel (fibrous by-products) by *Rhizopus* sp. The authors reported that over 35 of the original cellulose content of the substrate was lost during the solid-state fermentation. *A. niger* grown on rye-grass straw (Han and Anderson, 1975) produced similar results as obtained in this study.

Fungi have the ability to produce a variety of enzymes. *A. niger, A. flavus* and *Penicilluim* sp. have been reported to be main sources of cellulase, amylase, hemicellulase, catalase, pectinase and xylanase (Hamlyn, 1998). These enzymes help to degrade the non-starch polysaccharides (NSPs) in the substrate to soluble sugar. Thus with the decrease in the amount of cellulose, a corresponding increase in the soluble sugar content is obtained. *A. niger*

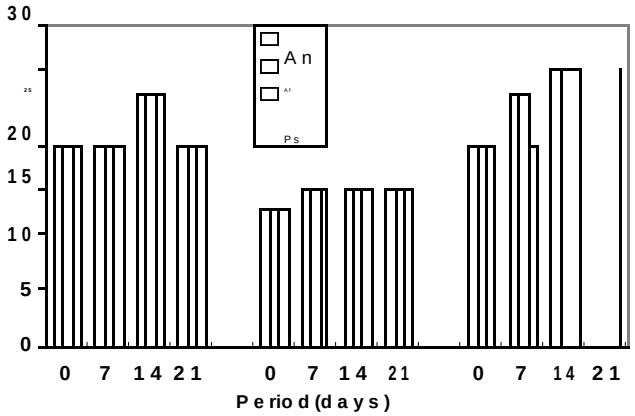

Figure 3. Changes in levels of crude protein in BDG, MO and WO biodegraded with *A. niger* (A n), *A. flavus* (A f) and *Penicillium* sp. (P s).

was able to bring about highest percentage in the cellulose reduction due to its more vigorous growth and therefore ability to produce more cellulolytic enzymes within a short period.

It is possible that synthesis and release of the enzymes are slowed down due perhaps to the changes in conditions in the medium. Ofuya and Nwanjiuba (1990) have reported that fungal enzyme controlled degradation responds to incubation time, pH and temperature of the medium. With fungal biomass increase, the nutrients in the substrate medium are quickly used up. Beyond 14 days therefore, the fungi start to take up the products of break down of the NSPs, hence the observed reduction in the sugar level. This means that 14 days is the optimum time for breakdown of NSPs in BDG, WO and MO using any of the three fungi.

The crude protein of the by-products also increased significantly ($P<0.05$). Percentage increases in BDG, MO and WO after 14 days were 31% 36% and 41% respectively with *A. niger;* 26%, 33% and 38% with *A. flavus* and 27%, 36% and 32% with *Penicillum* sp. Again beyond 14 days no significant increases were observed. *A. niger* produced the highest increase in protein in all the by-products. Similar results have been reported by Ofuya and Nwanjiuba (1990) when they cultured cassava peels with *Rhizopus* sp. The authors reported a 185% increase in the protein of the peels (from 5.6 to 16%). Such high increase than our results can be attributable to the fact that cassava peels are less fibrous than the BDG, MO and WO such that ease of degradability is more with cassava peels. Nevertheless, the ability of the fungi to take up products of polysaccharide degradation as demonstrated by reduced sugar levels after 14 days has been shown by results of the present study. The increase in protein levels obtained is as a result of the bioconversion of the sugars into mycelia protein. Solid state fermentation of biomass has been attempted as a means of elevating the total protein content by many workers (Reade and Gregory, 1975; Rodriguez et al., 1985; Dubresse et al., 1987). Balagopalan (1996) obtained similar results on fermentation of cassava products with *Trichoderma* sp.

In conclusion, results of the present study have demonstrated that the optimum period for the degradation of BDG, WO and MO by *A. niger, A. flavus* and *Peniciunim* sp. is 14 days. These fungi posses the capacity to degrade the non-starch polysaccharide contents of the agro-industrial by-products, converting them to simple sugars with a beneficial increase in energy and protein in these by-products. This raises the prospects of increase in the inclusion of these biodegraded by-products in poultry and pig diets which can help spare maize in the diets of these animals.

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