

Effects of Fungal Solid State Fermentation Using *Aspergillus niger* on the Nutritional Quality and Phytic Acid Content of Millet

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Abstract: The effect of solid state fermentation of *Aspergillus niger* on millet's nutritional quality and phytic acid content were investigated in this study. Ammonium sulphate and urea as Nitrogen sources were used in combination with millet at 10 gN kg⁻¹ substrate. The mixture was fermented for 84 h at 35°C and 90-95% RH in the laboratory. The results indicated that the crude protein content of millet was raised from 10.96-17.33%, while the crude lipid was lowered from 3.12-2.58%. The ash content value was greatly increased from 1.36-7.94%. Five percent changes in minerals (calcium, magnesium and phosphorus) content values were also observed. Phytic acid content value of 0.4 mg 100g⁻¹ reduced significantly to minimal 0.015 mg 100g⁻¹.

Key words: *Pennisetum americanum*, solid state fermentation, *Aspergillus niger*, nutritional quality, phytic acid content

INTRODUCTION

Recently, the use of millet (*Pennisetum americanum*) is becoming prominent in fish feed formulation as energy source because of its abundance and relatively low price. Millet is grown extensively around the world and is no doubt superior to other cereals with respect to some of the nutrients especially average protein, minerals and fat (Sharma and Kapoor, 1996), thereby suggesting that millet can be successfully used in fish feed formulation.

However, the inherently low protein, fat mineral contents and the presence of an anti-nutritional factor (phytic acid) affect the maximum utilization of the crop (Athapol *et al.*, 1992).

Fermentation processes using fungi are known to affect the chemical composition of food, improve the nutritive value and reduce the anti-nutritional factors (Sharma and Kapoor, 1996). According to Athapol *et al.* (1992), conversion of a part of the starch into protein by microbial fermentation has proved viable. Therefore, protein content of millet can be improved by solid state fermentation wherein protein is produced by microorganisms growing on millet without requiring aseptic conditions.

Solid state fermentation deals with the utilization of water-insoluble material for microbial growth and metabolism (Athapol *et al.*, 1992), moreover, it is usually carried out in semi-solid systems in the absence of free liquid.

Many microorganisms are capable of growing on solid substrate but only filamentous fungi can grow to a significant extent in the absence of free water.

Among the filamentous fungi, only Mucor, Rhizopy, *Aspergillus* and *Penicillium* have gained practical importance in solid state fermentation (Athapol *et al.*, 1992).

Especially, *Aspergillus niger* has been used in the fermentation of sweet potato root meal to increase the protein fatty acid mineral compositions and reduced the phytic acid content (Tewe *et al.*, 1999).

Presently, utilization of millet in fish feed is limited to raw millet meal, thus this study was proposed to investigate the removal of toxic

MATERIALS AND METHODS

Millet seeds were purchased at Bodija market in Ibadan and sterilized in an autoclave at 121°C for 30 min.

Test organism: *Aspergillus niger* was obtained from the culture collection of Microbiology laboratory, International Institute of Tropical Agriculture (IITA), Ibadan. The organism as were cultivated at 25°C on malt extract agar slants, containing (g L⁻¹): Lab malt extract agar 20.0; NO₂ 10.0; NaNO₃ 2.0; KH₄ 1.0; MgA0₄ 7H₂O 0.5; and sub-cultured every four weeks. The spores were harvest with a Tween 80 solution (10 mL, 0.01% v/v) which was then adjusted to give 10⁷ spores per mL with sterile water.

Inoculation technique: The sterilized Millet seeds served as substrate used for cultivating the fungus. The substrate was inoculated with minimal nutrient medium: Water containing the Nitrogen source (10 gN as ammonium sulphate and 10 gN as urea per kg substrate), Spores of *Aspergillus niger* and sulphuric acid to obtain an initial pH of 3.5-4.0. The inoculated Millet seeds were then spread on perforated wire mesh tray 1.5 inch in thickness and incubated at temperature and humidity fixed at 35°C and 95%, respectively for 84 h. At the end of the fermentation period, fermented Millet seeds were sundried for 48 h and ground in a hammer mill into fine powder to obtain the Millet meal. Samples were collected and analyzed for total protein, total lipid, minerals and phytic acid content at the end of experiment.

Ten gram Determination of pH of the wet sample was mixed with 100 mL of distilled water for 30-45 sec. The slurry was allowed to stand for 3 min before the pH was read using pH meter (Metler Toledo 320 pH meter).

Chemical analysis

Total protein: Crude protein content (N×6.25) of the sample was determined by the Micro-Kjedahl method (Abu and Tewe, 1996).

Lipid: Lipid content of the sample was assessed by the soxlet extraction method using petroleum ether (boiling point 40-60°C) in the electrothermal soxlet apparatus (Abu and Tewe, 1996).

Ash: Crude fibre content was determined according to AOAC (1990)

Mineral analysis: Concentrations of calcium, Potassium and Magnesium in the ash were determined by a Perkin Elmer-flame atomic absorption spectrophotometer.

Phytic acid: Phytic acid content was determined according to AOAC (1990).

RESULTS AND DISCUSSION

The changes in the crude protein, Crude lipid crude fibre and phytic acid contents of inoculated millet after 84 h of fermentation are shown in Table 1. The pH changed from 4.0- 3.5, which is within the range suitable for fungal growth. Abu and Tewe (Abu and Tewe, 1996) reported a pH range of 3.80-5.39 as suitable for growth of most fungi. The total protein was raised from 10.96-17.33% after fermentation. The results obtained in this study are comparable to those of Tewe *et al.* (1999).

Table 1: Proximate composition and phytic acid content of raw and fermented millet *Pennisetum americanum*

Component	Raw (%) mean value	Millet ±SD	Fermented (%) mean value	Millet ±SD
Crude Protein %	10.96	0.23	17.33	0.20
Crude Lipid %	3.12	0.02	2.58	0.20
Crude Fibre %	3.65	0.20	3.27	0.22
Phytic acid	0.70	0.02	0.02	0.02

Content (mg/100 g)
±SD. = Standard Deviation

Table 2: Mineral Composition of Raw and fermented Millet (*Pennisetum americanum*)

Component	Raw (%) mean value	Millet ±SD	Fermented (%) mean value	Millet ±SD
Calcium	0.03	0.01	0.11	0.02
Magnesium	0.059	0.50	0.091	0.01
Phosphorus	0.023	0.001	0.061	0.001

±SD. = Standard Deviation

Abu and Tewe (1996) and Abdallah *et al.* (1998) have reported up graded crude protein contents of sweet potatoes and cassava products, respectively. There is however, a decrease in total lipid level from 3.12-2.58% contrast to the report of Abu and Tewe (1996) in which total lipid level increased from substrate inoculated with *Aspergillus niger* for more than 72 h. Phytic acid content drastically reduced to insignificant level from 0.70-0.02 mg 100g⁻¹ and conforms with the report of Sharma and Kapoor (1996).

Other minerals not detected by flame atomic absorption spectroscopy

The mineral composition of raw and fermented millet are as presented in Table 2. Fermentation had relatively increased the calcium, magnesium and phosphorus. These observations are compared to the level found in the raw millet and similar to the reports of Tewe *et al.* (1999), where, the level of phytate was reduced significantly by fermentation thereby making some minerals available. This is also in line with the findings of Abdallah *et al.* (1998) which reported the effects of traditional processes on phytate and mineral contents of pearl millet.

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