

Physicochemical and proximate composition of mango (*Mangifera indica*) kernel cake fermented with mono-culture of fungal isolates obtained from naturally decomposed mango kernel

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Received June 28, 2008

Abstract

Ten fungal species were isolated from naturally decomposed mango kernel meal in the laboratory (NDMKML). They were identified as *Rhizopus oligosporus*, *Aspergillus niger*, *Rhizopus stolonifer*, *Penicillium chrysogenum*, *Cladosporium cladosporoides*, *Aspergillus flavus*, *Trichoderma longibrachiatum*, *Curvularia lunata*, *Sporendonema casei* and *Rhizopus oryzae*. The same fungal isolates were also obtained from naturally decomposed whole mango kernels that were collected on the farm (NDWMKF) except *Curvularia lunata*, *Rhizopus oligosporus* and *Rhizopus oryzae*. Solid state fermentation of mango kernel cake (MKC) using the fungal isolates in a mono-culture conditions showed significant ($P < 0.05$) reduction of the inherent anti-nutrients. The fungal isolates generally reduced the inherent anti-nutrients of the fermented MKCs to about half of the total contents of anti-nutrients in the unfermented MKC except the values of oxalate (77.41 mg/100 g) obtained for the *Aspergillus niger* fermented MKC that was higher than the values of unfermented MKM (54.24 mg/100 g) and the unfermented MKC (70.05 mg/100 g). There were significant ($P < 0.05$) changes in most of the valuable nutrients; such as crude protein, crude fibre, crude fat and glucose. The range of values obtained for the various nutrients of the mono-culture fermented MKC are crude protein (14.18% – 26.42%), crude fibre (1.16% – 5.03%), crude fat (3.24% – 6.78%), total ash (5.20% – 6.83%), glucose (16.54 mg/g – 24.69 mg/g) and total carbohydrates (48.54% – 61.28%). It is concluded that there may be improvement in nutrient bioavailability and utilization of MKC, if the fermented cake is used as a substitute for maize in poultry diets. [Life Science Journal. 2008; 5(4): 55 – 63] (ISSN: 1097 – 8135).

Keywords: Mango fruits; “Oori” cultivars; decomposed whole mango kernel; decomposed mango kernel meal; mango kernel cake; mono-culture fermentation

1 Introduction

One of the greatest challenges facing the livestock industry in the developing countries is the provision of nutritionally balanced and cost effective rations, since feed constitutes about 65% – 80% of the total cost of production. The situation has necessitated the need to source for an alternative or partial replacement of one of the most highly competitive feed ingredients such as

maize, which contributes about 60% – 80% of most formulated diets (Durunna *et al*, 2000; Abdulrashid *et al*, 2007).

Some of the unconventional feed resources that have been experimented include Africa locust bean meal (*Parkia filicoidea* welw), avocado seed meal (*Persea americana*), bambara groundnut meal (*Voandzeia subterranean*), coffee pulp meal (*Coffee arabica*), and mango seed kernel meal (*Mangifera indica*)(Joseph and Abolaji, 1997; Aregheore, 1998; Kperegbeyi and Onwumere, 2007).

However, most of these feeds contain anti-nutrients and toxic components such as saponins, lectins, tannins,

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trypsin inhibitors and cyanogenic glycosides which make them unsafe as protein and carbohydrate sources in livestock production (Aregheore, 1992). These anti-nutrients chelate divalent ions like Ca^{2+} , Mg^{2+} , Fe^{2+} , and Zn^{2+} and also react with the charged groups of protein and polysaccharides thereby forming indigestible complexes while the toxic substances interfere with nutrient bioavailability and utilization (Haslam, 1989; Reed, 1995; Giner-Chavez, 1996; Osagie, 1998).

Drying, soaking, leaching and fermentation have been reported to be simple means of detoxifying these feed sources to reduce the presence of the anti-nutrients and the toxic components (Aregheore, 1998). Traditional technologies that employ fungal fermentation include the production and flavouring of foods such as the production of miso, ragi and shoyu (soysuace) using *Aspergillus oryzae*, *Rhizopus* sp. and mixed-cultures of *Mucor* and *Rhizopus* sp respectively (Wainwright, 1992). Fermentation in this respect has largely been an uncontrolled chance inoculation. The consequence is the production of non uniform product by any standard. It is therefore imperative to develop unconventional alternative feed resources through careful identification, suitable fermentation processes and feeding trial evaluation of the abundant indigenous plant species for sustainable livestock production. The use of fungi in agricultural biotechnology is presently largely devoted to technologies aimed at treating unconventional tropical feed stuffs to inhibit or reduce some of their anti-nutritive factors, enhance nutrient bioavailability and utilization. Mango is a tropical plant which produces the mango fruit. In Nigeria, tones of the mango kernels are generated from the fruits annually as waste and thereby constituting environmental nuisance.

The objective of this work therefore, is to evaluate the physicochemical and proximate composition of mango kernel fermented with fungi under carefully controlled conditions with a view to determining their nutritive potentials in animal nutrition.

2 Materials and Methods

2.1 Source of mango fruits and processing of the seeds into mango kernel cake (MKC)

They were identified as "Oori" cultivars at the Ministry of Agriculture and Natural Resources (MANR) in Ilorin. Matured whole mango (*Mangifera indica*) fruits were purchased from some mango plantations in Ogbomoso town, Oyo State, Nigeria.

The peels and pulp were removed by washing in clean water, while the seeds were separated and cracked

manually to remove the shells and hulls. The kernels were dried in the oven (Gallenkamp) at $60 \pm 1^\circ\text{C}$ to constant weight after which it was ground by using a grinder. The resulting fine powder is referred to as the mango kernel meal (MKM).

The oil in the MKM was extracted with n-hexane by using a giant soxhlet extractor containing one kilogram (1 kg) of the kernel meal at a time for 3 hours to obtain the MKC.

2.2 Isolation and characterization of fungi from naturally decomposed mango kernel

A-glass fermenter was sterilized in the oven (Gallenkamp) at 160°C for 2 hours and then cooled to room temperature ($27 \pm 5^\circ\text{C}$).

One-kilogramme of MKM and one litre of sterile distilled water were poured into the sterile fermentor. The mash obtained was acidified with 10% tartaric acid to pH 3.5 (Oxoid manual, 1980) to suppress bacterial growth. This mixture was stirred properly to obtain a uniform mash, covered and incubated in the laboratory at room temperature ($27 \pm 5^\circ\text{C}$) for 5 days (Lawal *et al*, 2005). Fungi were isolated from the naturally decomposed MKM in the laboratory.

Naturally decomposed whole mango kernels were also collected into sterile specimen bottles on the farm before taken to the laboratory for fungal isolation. Fungal isolates were characterized and identified according to Harrow (1968); Samson and Von Reen-Hoekstra (1988).

2.3 Inoculation of MKCs for mono-culture fermentation

Fungal suspension of actively growing mid-log phase culture of each isolate was prepared according to the methods described by Sani *et al* (1992). One-kilogramme of autoclaved (sterile) MKC was mixed with 1 L of sterile distilled water in ten different fermenters and stirred properly to obtain a uniform mash. Twenty millilitres from each of the mono-culture suspension (5×10^4 spore/ml) was used as fermentation starter to inoculate each of the MKCs in the fermenters before the commencement of fermentation at ambient temperature ($27 \pm 5^\circ\text{C}$) for 168 hours (Lawal *et al*, 2005).

2.4 Determination of viable fungal counts

Total viable spores in the fermenting MKCs were determined by using serial dilution standard plate counts method (Jay, 1987).

2.5 Physicochemical analysis

The method of Rotter *et al* (1989) was used to assess the water soluble extractive (WSE), while water holding

capacity was determined according to the methods described by Zazueta-Murale *et al* (2006). Glucose was determined colorimetrically using DNSA method (Miller, 1959). The pH, titratable acidity and proximate analysis were carried out using standard methods (Egan *et al*, 1981; Cullison, 1982; AOAC, 1990).

2.6 Qualitative and quantitative analysis of the anti-nutrients

The anti-nutrients screening of fresh, dry and defatted mango kernel was carried out following the methods described by Odebisi and Sofowora (1978), Trease and Evans (1989). Phytate was quantitatively determined according to the method described by Wheeler and Ferrel (1971). Oxalate was determined using the method employed by Iwuoha and Kalu (1995), while tannin was determined using copper-acetate gravimetric method described by Joslyn (1970).

2.7 Statistical analysis

The experimental design used was a completely randomized design (CRD). Data obtained was analyzed by one-way ANOVA, while significant differences among the means were determined by using Duncan's New Multiple Range Test as outlined by Obi (2002).

3 Results and Discussion

Ten fungal isolates were identified to be associated with natural decomposition of MKM in the laboratory. The fungal isolates were *Rhizopus oligosporus*, *Aspergillus niger*, *Rhizopus stolonifer*, *Penicillium chrysogenum*, *Cladosporium cladosporoides*, *Aspergillus flavus*, *Trichoderma longibrachiatum*, *Curvularia lunata*, *Sporendonema casei* and *Rhizopus oryzae*. The same fungal isolates were also obtained from naturally decomposed whole mango kernels that were collected on the farm except *Curvularia lunata*, *Rhizopus oligosporus* and *Rhizopus oryzae* (Table 1).

The growths of fungal isolates in MKC during monoculture fermentation are shown in Figures 1 and 2. There was a rapid multiplication of the fungal isolates in the MKCs within the first 72 hours of fermentation as indicated by viable spore counts (Figure 1). Highest value of viable spores which ranged between \log_{10} 2.06 (115 spores) at zero hour and \log_{10} 3.40 (2512 spores) at 168 hours was recorded in the MKC fermented with *Aspergillus flavus* while, the lowest value of viable spores which also range between \log_{10} 1.12 (13 spores) at zero hour and \log_{10} 2.17 (148 spores) at 168 hours was obtained in the MKC containing *Rhizopus oligosporus*.

Table 1. Fungal isolates from naturally decomposed whole mango kernel and MKM

Fungal isolates	Colour/appearance on potato dextrose agar	Source of isolation	
		NDWMKF	NDMKML
<i>Rhizopus oligosporus</i>	brownish gray	-	+
<i>Aspergillus niger</i>	dark brown to black	+	+
<i>Rhizopus stolonifer</i>	grayish brown	+	+
<i>Penicillium chrysogenum</i>	bluish-green with broad white margin	+	+
<i>Cladosporium cladosporoides</i>	brown or grayish green	+	+
<i>Aspergillus flavus</i>	yellowish green	+	+
<i>Trichoderma longibrachiatum</i>	whitish turning orange to red	+	+
<i>Curvularia lunata</i>	dark gray	-	+
<i>Sporendonema casei</i>	whitish turning orange to red	+	+
<i>Rhizopus oryzae</i>	whitish turning brownish gray	-	+

+: present; -: absent; NDWMKF: naturally decomposed whole mango kernel on the farm; NDMKML: naturally decomposed MKM in the laboratory.

during the fermentation period.

Generally, the fermenting MKCs showed increase in temperature with increase in fermentation period (Table 2). This is expected since fermentation is an exothermic reaction. The internal temperature of the fermented MKCs was significantly higher ($P < 0.05$) at 72 to 168 hours than the unfermented MKC (Table 2). The values of internal temperature of all the fermented MKCs were 26.4 °C at zero hour. Although, the *Aspergillus flavus* fermented MKC and the *Penicillium chrysogenum* fermented MKC had the highest internal temperature of 34.4 °C respectively, while the *Sporendonema casei* fermented MKC had the lowest internal temperature (32.3 °C) at 168 hours when the experiment was terminated.

There was a general decrease in pH of the MKCs during the fermentation period (Table 3). The range of pH of the fermenting MKCs falls between 3.75 and 4.66 after 168 hours. These values were significantly lower ($P < 0.05$) than the pH of 5.12 obtained for the unfermented MKC. The titratable acidity of all the fermented MKCs also increased significantly ($P < 0.05$) when compared with the unfermented MKC (Table 4). Values obtained for the fermented MKCs range between 13.14 mg/100 g and 18.67 mg/100 g at 168 hours.

The gradual decrease in pH with the resultant

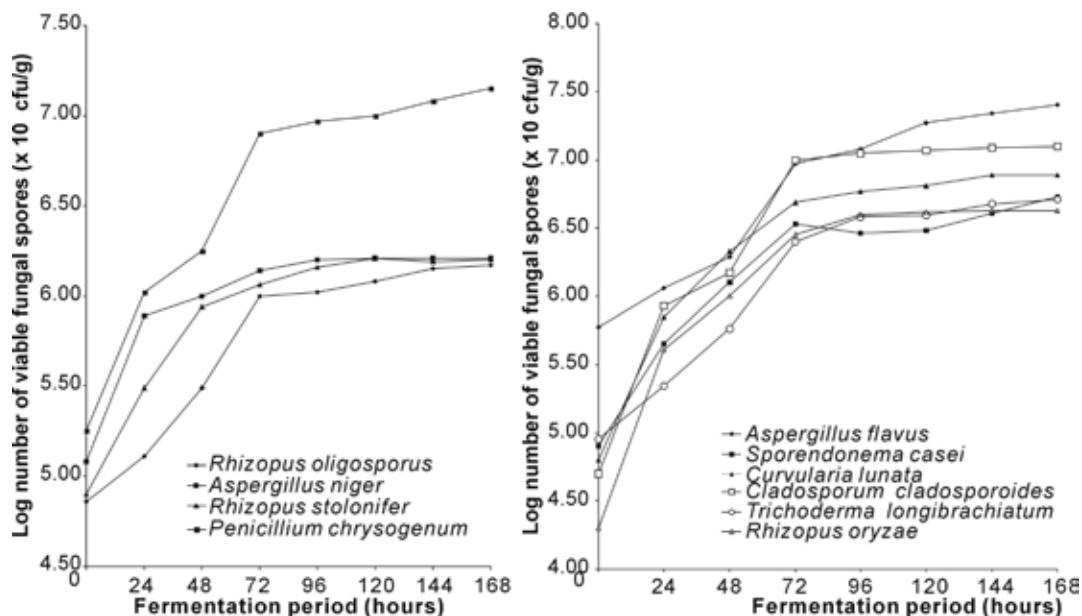


Figure 1. Growth of fungal isolates in MKC during mono-culture fermentation.

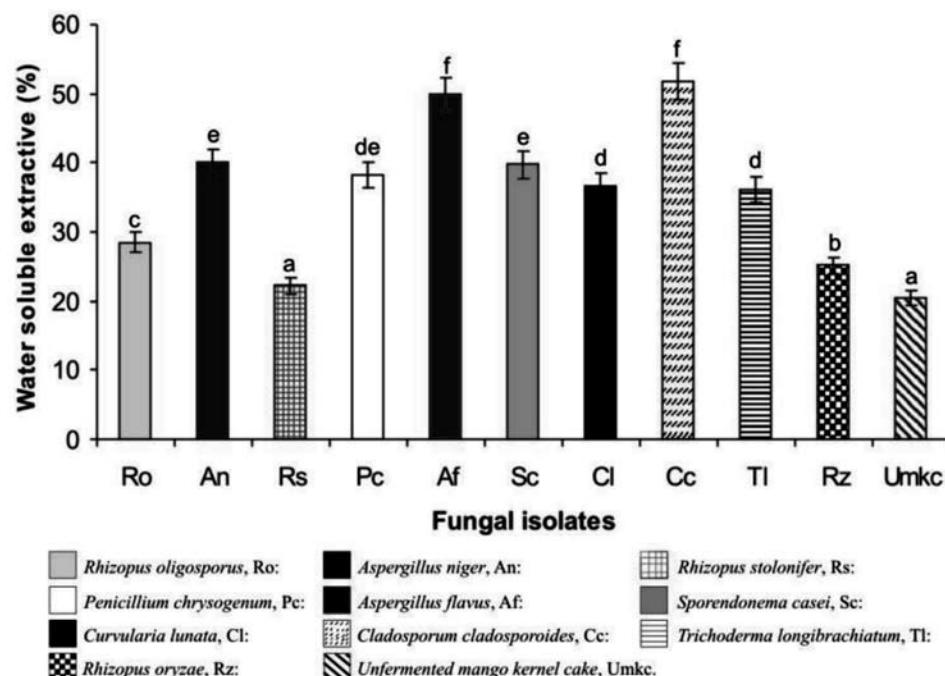


Figure 2. Effect of mono-culture fermentation at ambient temperature on water soluble extractive in MKC.

increase in titratable acidity may be attributed to the rapid growth of the fungal isolates in the fermenting MKCs and the concurrent production of some metabolic substances, such as organic acids since fermentation was mostly aerobic. The water soluble extractives of the fermented MKCs increased significantly ($P < 0.05$) when

compared with the unfermented MKC (Figure 2). The highest (51.74%) was in *Cladosporum cladosporoides* fermented MKC and least (22.20%) in *Rhizopus stolonifer* fermented MKC. This may be due to the breakdown of some complex molecules into simpler organic molecules by the fungal isolates which resulted in the

Table 2. Effect of mono-culture fermentation on internal temperature of the fermenting MKC

Fungal isolates	Fermentation period (hours)/temperature (°C)							
	0	24	48	72	96	120	144	168
<i>Rhizopus oligosporus</i>	26.4 ^a	25.0 ^a	28.4 ^a	29.5 ^{abc}	32.4 ^b	32.5 ^b	33.8 ^b	32.5 ^b
<i>Aspergillus niger</i>	26.4 ^a	25.5 ^a	28.6 ^a	30.1 ^{abc}	32.5 ^b	33.5 ^b	32.5 ^b	33.7 ^b
<i>Rhizopus stolonifer</i>	26.4 ^a	26.3 ^a	27.5 ^a	31.2 ^{bc}	32.0 ^b	33.7 ^b	33.5 ^b	33.5 ^b
<i>Penicillium chrysogenum</i>	26.4 ^a	24.5 ^a	26.1 ^a	32.4 ^c	33.5 ^b	32.5 ^b	33.5 ^b	34.4 ^b
<i>Aspergillus flavus</i>	26.4 ^a	25.5 ^a	26.5 ^a	30.5 ^{abc}	33.0 ^b	32.9 ^b	34.3 ^b	34.4 ^b
<i>Sporendonema casei</i>	26.4 ^a	24.5 ^a	24.5 ^a	25.5 ^a	29.3 ^{ab}	32.3 ^b	32.1 ^b	32.3 ^b
<i>Curvularia lunata</i>	26.4 ^a	25.2 ^a	26.0 ^a	33.3 ^c	32.2 ^b	33.5 ^b	34.1 ^b	33.5 ^b
<i>Cladosporium cladosporoides</i>	26.4 ^a	25.5 ^a	25.5 ^a	31.0 ^{bc}	33.8 ^b	31.6 ^b	32.5 ^b	33.5 ^b
<i>Trichoderma longibrachiatum</i>	26.4 ^a	25.1 ^a	27.0 ^a	29.7 ^{abc}	32.9 ^b	33.4 ^b	33.6 ^b	33.5 ^b
<i>Rhizopus oryzae</i>	26.4 ^a	26.1 ^a	27.5 ^a	31.4 ^{bc}	33.3 ^b	33.4 ^b	34.1 ^b	33.0 ^b
Unfermented cake	26.4 ^a	26.4 ^a	25.9 ^a	26.3 ^{ab}	26.3 ^a	26.7 ^a	26.7 ^a	26.5 ^a
SEM	0.00	1.64	1.56	1.67	1.46	0.83	1.54	1.66

Values are means of four replicates readings. SEM: standard error of mean; ^{a,b,c}: means on the same column followed by different superscripts differ significantly ($P < 0.05$).

Table 3. Changes in the pH of MKC during mono-culture fermentation at ambient temperature

Fungal isolates	Fermentation period (hours)/pH							
	0	24	48	72	96	120	144	168
<i>Rhizopus oligosporus</i>	5.12 ^a	4.95 ^a	4.80 ^b	4.53 ^b	4.40 ^b	4.27 ^b	4.18 ^b	4.09 ^b
<i>Aspergillus niger</i>	5.12 ^a	4.94 ^a	4.79 ^b	4.40 ^a	4.08 ^a	3.92 ^a	3.75 ^a	3.75 ^a
<i>Rhizopus stolonifer</i>	5.12 ^a	4.96 ^a	4.80 ^b	4.68 ^{cd}	4.64 ^f	4.64 ^{ef}	4.63 ^g	4.62 ^g
<i>Penicillium chrysogenum</i>	5.12 ^a	5.01 ^b	4.79 ^b	4.69 ^{cd}	4.68 ^f	4.67 ^f	4.67 ^g	4.66 ^h
<i>Aspergillus flavus</i>	5.12 ^a	4.96 ^a	4.70 ^a	4.68 ^{cd}	4.68 ^f	4.68 ^f	4.66 ^g	4.66 ^h
<i>Sporendonema casei</i>	5.12 ^a	5.05 ^a	4.79 ^b	4.73 ^{de}	4.64 ^f	4.51 ^{cd}	4.45 ^e	4.42 ^c
<i>Curvularia lunata</i>	5.12 ^a	5.09 ^a	4.80 ^b	4.65 ^c	4.58 ^e	4.55 ^{de}	4.53 ^f	4.52 ^f
<i>Cladosporium cladosporoides</i>	5.12 ^a	5.02 ^b	4.82 ^b	4.75 ^e	4.45 ^c	4.41 ^c	4.26 ^c	4.23 ^c
<i>Trichoderma longibrachiatum</i>	5.12 ^a	4.96 ^a	4.78 ^b	4.68 ^{cd}	4.53 ^d	4.51 ^{cd}	4.43 ^e	4.38 ^d
<i>Rhizopus oryzae</i>	5.12 ^a	4.96 ^a	4.79 ^b	4.69 ^{cd}	4.53 ^d	4.49 ^{cd}	4.36 ^d	4.38 ^d
Unfermented cake	5.12 ^a	5.12 ^a	5.11 ^b	5.11 ^f	5.11 ^g	5.12 ^g	5.12 ^h	5.12 ⁱ
SEM	1.74	0.92	0.60	1.70	1.55	1.03	0.74	1.23

Values are means of three replicates reading. SEM: standard error of mean; ^{a,b,c,d,e,f,g,h,i}: means within column having different superscripts differ significantly ($P < 0.05$).

Table 4. Effect of mono-culture fermentation at ambient temperature on titratable acidity of MKC

Fungal isolates	Fermentation period (hours)/titratable acidity (mg/100 g)							
	0	24	48	72	96	120	144	168
<i>Rhizopus oligosporus</i>	10.61 ^a	11.10 ^{cd}	12.93 ^d	13.58 ^e	13.68 ^c	15.15 ^c	15.44 ^c	15.45 ^c
<i>Aspergillus niger</i>	10.61 ^a	10.39 ^a	10.45 ^b	12.14 ^{cd}	14.36 ^{cd}	16.13 ^d	16.14 ^c	16.62 ^d
<i>Rhizopus stolonifer</i>	10.61 ^a	10.61 ^{ab}	6.91 ^a	10.12 ^a	12.71 ^b	12.93 ^b	13.19 ^b	13.14 ^b
<i>Penicillium chrysogenum</i>	10.61 ^a	10.75 ^{abcd}	10.32 ^b	11.20 ^{bc}	11.62 ^b	17.16 ^e	17.16 ^e	17.27 ^e
<i>Aspergillus flavus</i>	10.61 ^a	12.79 ^e	14.35 ^e	14.93 ^f	15.72 ^f	17.09 ^e	17.16 ^f	17.27 ^e
<i>Sporendonema casei</i>	10.61 ^a	10.89 ^{bcd}	11.66 ^c	13.70 ^e	15.55 ^{ef}	16.05 ^d	15.82 ^{cd}	16.33 ^d
<i>Curvularia lunata</i>	10.61 ^a	11.20 ^d	12.76 ^d	13.08 ^{de}	15.05 ^{def}	15.94 ^d	16.15 ^{de}	16.45 ^d
<i>Cladosporium cladosporoides</i>	10.61 ^a	10.89 ^{bcd}	10.89 ^{bc}	11.89 ^c	14.71 ^{de}	15.27 ^c	15.53 ^d	16.24 ^d
<i>Trichoderma longibrachiatum</i>	10.61 ^a	10.73 ^{abc}	10.91 ^{bc}	15.48 ^f	17.10 ^g	17.42 ^e	18.00 ^g	18.67 ^f
<i>Rhizopus oryzae</i>	10.61 ^a	10.63 ^{ab}	10.59 ^b	13.55 ^e	16.85 ^g	17.32 ^e	17.34 ^f	17.36 ^e
Unfermented cake	10.61 ^a	10.60 ^{ab}	10.60 ^b	10.60 ^{ab}	10.58 ^a	10.61 ^a	10.62 ^a	10.61 ^a
SEM	0.00	1.43	0.32	0.34	0.32	0.23	0.20	1.34

Values are means of three replicates readings. SEM: standard error of mean; ^{a,b,c,d,e,f,g}: means within column having different superscripts differ significantly ($P < 0.05$).

observed increase in the water soluble extractive.

The water holding capacity ($1.65 \text{ g H}_2\text{O/g dried cake}$) of the unfermented MKC is significantly lower ($P < 0.05$) than the values obtained for the fermented MKCs (Figure 3). The highest value ($2.49 \text{ g H}_2\text{O/g dried cake}$) was in *Rhizopus stolonifer* fermented MKC and the least ($1.94 \text{ g H}_2\text{O/g dried cake}$) was obtained in the *Cladosporium cladosporoides* fermented MKC. The higher values recorded among other factors may be due to the fibre contents of the fermented MKCs and the breakdown of starch into simpler organic sugars which has higher capacity for holding water.

The proximate composition of the fermented MKCs is shown in Table 5. There was an increase in valuable nutrients: such as mineral ash, crude protein, dietary fibre and glucose. The range of values obtained for the nutrients in the fermented MKCs are crude protein ($14.18\% - 26.42\%$), crude fibre ($1.16\% - 5.03\%$), total ash ($5.20\% - 6.83\%$) and glucose ($16.54\% - 24.69 \text{ mg/g}$). These values were mostly significantly higher ($P < 0.05$) in comparison with the unfermented MKC that contained crude protein (15.56%), crude fibre (2.64%), total ash (2.63%) and glucose (15.12 mg/g). On the contrary, the range of values obtained for crude fat ($3.24\% - 6.78\%$) and total carbohydrates ($48.54\% - 61.28\%$) of the fermented MKCs are significantly lower ($P < 0.05$) than the values (crude fat: 6.98% and total carbohydrates:

64.84%) of the unfermented MKC. The nutrients in the fermented MKCs may be of advantage to animals for feeding purpose.

The hydrolysis of carbohydrates in the fermented MKCs which produces glucose may have been aided by the fungal enzymes such as endocarbohydrases or exocarbohydrases and β -D-glucosidase since there was increase in the glucose content of all the fermented MKCs after 168 hours of mono-culture fermentation (Table 5). Fungal glucoamylase and α -amylases have been shown to be produced by *Aspergillus niger*, *Rhizopus oryzae* and some other filamentous fungi (Sani *et al*, 1992; Wainwright, 1992). These enzymes may have been responsible for the hydrolysis and conversion of starch to glucose in the MKC. The reduction in the fat of the mono-culture fermented MKCs may have been due to the lipolytic activity of fat decomposing enzyme (lipase) produced by some fungi vis-à-vis the use of the products of hydrolysis for the synthesis of fungal cell constituent and growth. The noticeable increase in crude protein in the mono-culture fermented MKCs could be attributed to the addition of mycoprotein (single cell protein) which may be due to the addition of non-protein nitrogen from amide and nucleic acid synthesis by fungal cells. The slight increase in crude fibre may be due to the production of some dietary fibre-rich substances as component parts of mycoprotein deposited in the

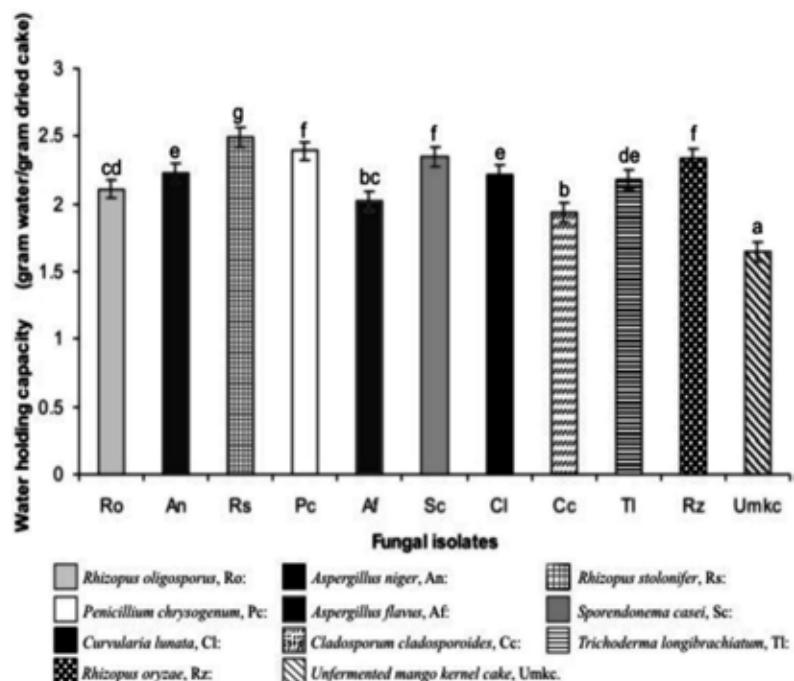


Figure 3. Effect of mono-culture fermentation at ambient temperature on water soluble extractive in MKC.

Table 5. Proximate composition of MKC fermented with mono-culture of fungal isolates at ambient temperature

Fungal isolates	Proximate composition (%)						
	Moisture content	Total ash	Crude fat	Crude protein	Crude fibre	NFE (CHO)	Glucose (mg/g)
<i>Rhizopus oligosporus</i>	9.14 ^{cde}	5.20 ^b	4.39 ^{ab}	26.42 ^f	3.16 ^{bcd}	50.69 ^b	16.54 ^{ab}
<i>Aspergillus niger</i>	10.50 ^e	5.46 ^{bc}	3.24 ^a	23.89 ^{ef}	4.30 ^{de}	53.61 ^b	19.68 ^{bcd}
<i>Rhizopus stolonifer</i>	7.97 ^{abcd}	6.62 ^{bc}	6.78 ^d	19.88 ^{cd}	1.16 ^a	57.59 ^c	17.09 ^{ab}
<i>Penicillium chrysogenum</i>	8.29 ^{abcd}	6.10 ^{bc}	5.47 ^{bc}	22.90 ^{de}	3.82 ^{cde}	52.42 ^b	20.23 ^{bcd}
<i>Aspergillus flavus</i>	8.81 ^{bcd}	9.17 ^d	6.03 ^{cd}	22.42 ^{de}	5.03 ^e	48.54 ^a	18.37 ^{ab}
<i>Sporendonema casei</i>	7.94 ^{abcd}	5.55 ^{bc}	5.29 ^{bc}	18.04 ^{bc}	3.04 ^{bcd}	60.14 ^c	22.09 ^{cde}
<i>Curvularia lunata</i>	8.05 ^{abcd}	6.37 ^{bc}	4.98 ^{bc}	17.64 ^{bc}	3.36 ^{bcd}	59.60 ^c	23.30 ^{de}
<i>Cladosporium cladosporoides</i>	9.29 ^{de}	6.72 ^c	3.50 ^a	14.18 ^a	5.03 ^e	61.28 ^c	24.69 ^e
<i>Trichoderma longibrachiatum</i>	7.48 ^{abc}	6.07 ^{bc}	3.58 ^a	18.38 ^{bc}	5.01 ^e	59.48 ^c	19.27 ^{bcd}
<i>Rhizopus oryzae</i>	7.00 ^a	6.83 ^c	5.57 ^{bcd}	17.74 ^{bc}	2.20 ^{ab}	60.66 ^c	20.18 ^{bcd}
Unfermented cake	7.35 ^{ab}	2.63 ^a	6.98 ^d	15.56 ^{ab}	2.64 ^{bc}	64.84 ^d	15.12 ^a
SEM	0.49	0.42	0.38	1.03	0.37	0.99	1.07

Values are means of three replicates determinations. NFE: nitrogen free extract; ^{a,b,c,d,e,f} means on the same column followed by different superscripts differ significantly ($P < 0.05$); SEM: standard error of mean.

fermented MKCs. The final product of a well processed mycoprotein food was reported to have 44.3% protein, 18.3% dietary fibre and 13.0% fat (Trinci, 1991).

The anti-nutrients in MKM and MKC are shown in Table 6. They include tannins, flavonoids, phenolic compound, oxalates and phytates. They were not removed by drying or defatting. The presence of tannins, oxalates, phytate and other anti-nutrients in the unfermented MKCs after drying treatment indicated that the anti-nutrients are heat stable (Table 6). Similar observation has been reported in legume seeds by Apata (1990).

The quantitative analysis of anti-nutrients in the mono-culture fermented MKCs revealed a significant ($P < 0.05$) decrease when compared with the unfermented MKC (Table 7). The fungal isolates generally reduced the inherent anti-nutrients of the fermented MKCs to about half of the total contents of anti-nutrients in the unfermented MKC except the values of oxalate (77.41 mg/100 g) obtained for the *Aspergillus niger* fermented MKC that was higher than the values of unfermented MKM (54.24 mg/100 g) and the unfermented MKC (70.05 mg/100 g). The range of values obtained for the anti-nutrients in the fermented MKCs are tannins (52.3 – 91.3 g/kg), phytate (0.18 – 0.64 mol/kg) and oxalates (7.51 – 28.62 mg/100 g). High intakes of plants rich in tannins and oxalates have been reported to be poisonous to human and animals (Apata, 1990). Phytate has been shown to be involved in the complexing of dietary essential minerals in legumes and cereals, thus rendering

them poorly available to monogastric animals (Oluremi *et al*, 2007).

Table 6. Effect of drying (60 °C) and defatting on the qualitative assessment of anti-nutrients in mango kernel

Mango kernel (processed/unprocessed)	Fresh kernel (undried)	Dried MKM	Dried MKC
Saponin	–	–	–
Tannin	+	+	+
Steroids	–	–	–
Flavonoid	+	+	+
Phenolic compound	+	+	+
Cyanogenic glycosides	–	–	–
Alkaloids	–	–	–
Oxalates	+	+	+
Phytate	+	+	+
Triterpene	–	–	–

+: detected; -: not detected.

4 Conclusion

In conclusion, the mono-culture of fungal isolates associated with the naturally decomposed whole mango kernel could be used under controlled solid state fermentation conditions to improve on the nutritional quality of MKC. The level of anti-nutrients in each of the fungal mono-culture fermented MKC may be safe for

animal consumption and feeding some of the fermented MKCs as supplement for maize in poultry diets may results in improvement of nutrients bioavailability and utilization over the unfermented MKC.

Table 7. Effect of mono-culture fermentation at ambient temperature ($27 \pm 5^\circ\text{C}$) on the anti-nutrients in MKC

Fungal isolates	Anti-nutrients		
	Tannin (g/kg)	Phytate (mol/kg)	Oxalates (mg/100 g)
<i>Rhizopus oligosporus</i>	58.7 ^b	0.61 ^c	27.33 ^e
<i>Aspergillus niger</i>	52.3 ^a	0.64 ^c	77.41 ^h
<i>Rhizopus stolonifer</i>	71.0 ^{cd}	0.38 ^b	16.91 ^e
<i>Penicillium chrysogenum</i>	67.7 ^c	0.61 ^c	10.25 ^a
<i>Aspergillus flavus</i>	73.6 ^d	0.18 ^a	14.09 ^b
<i>Sporendonema casei</i>	91.3 ^f	0.37 ^b	23.06 ^d
<i>Curvularia lunata</i>	68.0 ^c	0.38 ^b	19.65 ^e
<i>Cladosporum cladosporoides</i>	56.0 ^{ab}	0.40 ^b	7.51 ^a
<i>Trichoderma longibrachiatum</i>	83.7 ^e	0.21 ^a	28.62 ^e
<i>Rhizopus oryzae</i>	70.3 ^{cd}	0.59 ^c	19.22 ^c
Unfermented meal (umkc)	157.3 ^g	0.99 ^d	54.24 ^f
Unfermented cake (umkc)	164.5 ^h	1.34 ^e	70.05 ^g
SEM	1.59	1.03	1.09

Values are means of three replicates determinations. SEM: standard error of mean; ^{a,b,c,d,e,f,g,h}: means within column having different superscripts differ significantly ($P < 0.05$).

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