African Journal of Biotechnology Vol. 3 (1), pp. 52-59, January 2004
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2004 Academic Journals

Full Length Research Paper

Effect of lemon grass (*Cymbopogon citratus* Stapf) powder and essential oil on mould deterioration and aflatoxin contamination of melon seeds (*Colocynthis citrullus* L.)

S.A. BANKOLE[1*] and A.O. JODA[2]

[1]Department of Biological Sciences, Olabisi Onabanjo University, PMB 2002, Ago-Iwoye, Ogun State, Nigeria.
[2]Department of Crop Production, Olabisi Onabanjo University, PMB 2002, Ago-Iwoye, Ogun State, Nigeria.

Accepted 13 November 2003

Experiments were carried out to determine the potential of using the powder and essential oil from dried ground leaves of Cymbopogon citratus (lemon grass) to control storage deterioration and aflatoxin contamination of melon seeds (Colocynthis citrullus L.). Four mould species: Aspergillus flavus, A. niger, A. tamarii and Penicillium citrinum were inoculated in the form of conidia suspension (approx. 10^6 conidia per ml) unto shelled melon seeds. The powdered dry leaves and essential oil from lemon grass were mixed with the inoculated seeds at levels ranging from 1-10% (w/w) and 0.1 to 1%v/vt respectively. The ground leaves significantly reduced the extent of deterioration in melon seeds inoculat4ed with different fungi compared to the untreated inoculated seeds. The essential oil at 0.1 and 0.25% (v/w) and ground leaves at 10% (w/w) significantly reduced deterioration and aflatoxin production in shelled melon seeds inoculated with toxigenic A. flavus. At higher dosages (0.5 and 1.0% v/w), the essential completely prevented aflatoxin production. After 6 months in farmers' stores, unshelled melon seeds treated with 0.5% (v/w) of essential oil and 10% (w/w) of powdered leaves of C. citratus had significantly lower proportion of visibly diseased seeds and Aspergillus spp infestation levels and significantly higher seed germination compared to the untreated seeds. The oil content, free fatty acid and peroxide values in seeds protected with essential oil after 6 months did not significantly differ from the values in seed before storage. The efficacy of the essential oil in preserving the quality of melon seeds in stores was statistically at par with that of fungicide (iprodione) treatment.

Key words: Aflatoxin, Cymbopogon citratus, essential oil, powdered leaves, melon seeds, mould deterioration, oil content, free fatty acid, peroxide values, seed germination.

INTRODUCTION

Melon seeds (Colocynthis citrullus L.) are very important as a condiment in Nigerian local soup. They contain over 50% oil, 28% protein (60% in the defatted meal), 2.50% soluble sugars and 11% starch (Oyolu, 1977). Thus, melon seeds constitute a very valuable source of oil and protein. Melon seeds are consumed in various forms such as 'egusi' soup, melon ball snacks and ogiri (fermented melon seed condiment used in seasoning). Some rural inhabitants of southeastern Nigeria mix milled melon seeds with the ground fungus Pleurotus tuber regium, and mould them into stabilized balls to substitute meat in their diet (Nwokolo and Sim, 1987).

Fungi of the genus Aspergillus and Penicillium are widely distributed storage fungi of melon seeds, causing

*Corresponding author. Email: sabankole@juno.com.

seed discolourations, decreased nutritive value, increase in free fatty acid and peroxide values, decreased seed germination and producing a number of toxic metabolites including aflatoxin (Aboaba and Amasike, 1991; Bankole, 1993; Bankole et al., 1999). Aflatoxins, the toxic metabolites produced by Aspergillus flavus and Aspergillus parasiticus were detected in over 30% of melon seed samples from markets and stores in Nigeria (Bankole and Adebanjo, 2004). Naturally occurring mixtures of aflatoxins and aflatoxin B_1 have been classified as Class 1 human carcinogens (IARC, 1993). Aflatoxins have been associated with elevated rate of liver cancer, growth stunting and immunotoxicity in West Africa (Gong et al., 2002; Turner et al., 2003). Based on concern of the hazards to livestock and man, concerted effort is now being directed at finding very cheap and reliable methods of minimizing aflatoxin formation in stored commodities (Bankole and Adebanjo, 2003).

Fungicides such as Iprodione and Aliette have been found to be effective against mould deterioration of melon seeds in stores (Bankole, 1998). However, fungicides have various setbacks such as development of resistant strains in the treated fungi, environmental toxic residues and toxicity to consumers. Furthermore, many Nigerian farmers find fungicides too expensive or not available at the appropriate time. Consequently, farmers resort to selling the melon seeds at low price immediately after harvesting and sun drying or mix the seeds with plant materials before loading them into stores. The present study was designed to evaluate the activity of dry powdered leaves and essential oil from *Cymbopogon citratus* against mould deterioration and aflatoxin production in melon seeds.

MATERIALS AND METHODS

Collection of plant materials

Leaves of *Cymbopogon citratus* were collected in Yewa part of Ogun State, Nigeria. The identity of the plant was confirmed by botanists of the Department of Biological Sciences, Olabisi Onabanjo University Herbarium. They were air-dried under the shade (25-29^{0}C) until the leaves became crispy dry after 5 days. The dried leaves were powdered in a coffee grinder, and sieved with a 0.5 mm size mesh.

Extraction of essential oil

The components of half of the dried leaves were extracted for the essential oil as follows. Two hundred and fifty grams of the powdered leaves was put in a round bottom flask, 1000 ml of distilled water was added and then subjected to hydro distillation in a modified Clevenger apparatus for 8 hours (Bankole, 1997). The oil recovered was dried over anhydrous sodium sulphate and kept in the refrigerator at 4^{0}C before use.

Sources of test fungi and preparation of conidia suspension

The test fungi: *Aspergillus flavus*, *A. niger*, *A. tamarii* and *Penicillium citrinum* were isolated from post harvest melon seeds (Bankole et al., 1999). Pure cultures of the fungi were maintained on potato dextrose agar complemented with 60 µg/ml chloramphenicol in the culture collection centre of the Department of Biological Sciences, Olabisi Onabanjo University, Ago-Iwoye. The aflatoxigenic *A. flavus* OG-028 used was isolated from maize grains (Bankole, 1994) and was found to be a high aflatoxin producing strain (Bankole, 1997).

The preparation of conidia suspension was by flooding the surfaces of 7-10 day old cultures of fungi on plates with 10 ml of 0.05% Tween 80 in sterile distilled water, gently rubbing the surfaces with bent glass rod to dislodge the spores, and filtering the suspension twice through two folds of cheese cloth to remove mycelia fragments. The conidia suspensions were pelleted by centrifugation at 2000 rpm for 3 min and the pellets were resuspended in 0.05% Tween 80 in sterile distilled water .The concentration of each fungus was adjusted to approximately 10^6 conidia/ml by dilution and counting with a haemocytometer (Hawksley BS748).

Melon seed sample

The most popular melon seed variety 'Bara' with black edges was purchased shortly after harvesting and sun drying from a local farmer in Imeko, Ogun State, Nigeria. Half of the seeds were hand peeled to remove the shells, and the discoloured and visibly mouldy seeds were removed. The seeds were surface sterilized in 1% NaOCl for one min, followed by three successive rinses in sterile distilled water. The seed moisture content which was 8.8% was equilibrated to approximately 14% by the addition of sterile distilled water.

Bioassay with powdered leaves of *C.citrates*

Five hundred grams of shelled melon seeds were distributed in each of several tightly stoppered glass jars, twenty ml of conidia suspension of each fungus was introduced into the jars in triplicates and then mixed with 5, 10, 25 or 50 g of fine powder of leaves corresponding to concentrations of 1.0, 2.0, 5.0 and 10.0% (w/w). The control jars contained the melon seeds and conidia suspension without any powdered leaves. The jars were incubated under room conditions (28±2^{0}C) with manual shaking done twice daily. After 14 days, the effect of the powdered leaves on the mould deterioration of melon seeds was assessed by determining the oil content, free fatty acid content and the peroxide values. The oil content was determined by drying and extraction for 6 h in a Soxhlet apparatus with petroleum spirit (boiling point, 40-60^{0}C) (Kuku and Adeniji, 1976). The free fatty acid content of the extracted oil was determined titrimetrically by the method of Ogundero (1981), while the peroxide values were determined using the method of Pearson (1970).

Bioassay with essential oil and powdered leaves

Fifty grams of shelled melon seeds were distributed in each of several conical flasks and each was inoculated with 5 ml conidia suspension of the toxigenic *A. flavus*. Different dosages of the oil were added to flasks in triplicates to give concentrations of 0.1, 0.25, 0.5 and 1% (v/w). Powdered leaves at the rate of 10% (w/w) was introduced into another set of flasks containing inoculated seeds. The control was also similarly set up but without the powdered leaves or essential oil.

Following incubation under room conditions for 14 days, the content of each flask was analysed for oil content, free fatty acid content and peroxide values as previously described. The aflatoxin B_1 level in each flask was determined by the method of Bankole et

Table 1. Effect of dry ground leaves of *C. citratus* on the oil content of melon seeds inoculated with different fungi.

Fungus	Dosage (wt/wt)					
	0	**1.0**	**2.0**	**5.0**	**10.0**	**LSD**
A. flavus	43.5	44.3	45.8	49.7	52.7	5.6
A. niger	46.2	46.7	48.5	50.3	51.4	3.3
A. tamarii	45.3	46.6	47.7	49.5	51.8	3.9
P.citrinum	46.5	47.2	49.8	51.5	52.3	2.6
LSD	NS	NS	NS	NS	NS	

NS = No significant difference.

al. (1996). The content of each container was extracted with 7:3 methanol-water, analysed by thin layer chromatography using 90:30:2 (v:v:v) toluene/isoamylalcohol/methanol, and an aflatoxin B_1 standard (Sigma). The identity of aflatoxin was confirmed by derivatization with trifluoroacetic acid (Stack and Pohland, 1975). The detection limit was 5 ng/g.

Storage trials in farmers' stores

Melon seeds that were not shelled with a moisture content of 8.8% were divided into four lots of 5 kg each. One lot was treated with essential oil to give 0.5% (v/w). The second lot was treated with powdered leaves at the rate of 10% (w/w). The third lot was treated with fungicide, iprodione (Rovral 50%a.i. Rhone Poulenc) at the rate of 0.3% (w/w) (the level of iprodione found to be effective against storage deterioration of melon seeds; Bankole, 1998). The fourth lot was the untreated control. Each lot was packed in woven polypropylene sacks (the type of storage bag commonly used by farmers in Nigeria) and each treatment was replicated 3 times. The sacks were kept in farmers' stores at three locations in a randomized block design. The locations were Mamu (Ijebu North Local Government, Ogun State), Oyero (Owode Local Government, Ogun State) and Epe (Epe Local Government, Lagos State). Samples were drawn from each sack immediately after bagging and on a monthly basis for 6 months using the method of Fan et al. (1976) by shaking each bag before taking 300 g seeds. At each sampling, the proportion of visibly diseased seeds, *Aspergillus* infection and seed germination were evaluated. The percentage incidence of diseased seeds was determined by scoring for the presence of visibly mouldy, rotted or discoloured seeds in 200 seeds per sack.

To determine *Aspergillus* infection, 100 seeds were surface sterilized by immersion in 1% NaOCl for one minute and rinsed thrice with sterile distilled water, then ten seeds were placed on each of 10 plates of potato dextrose agar plus chloramphenicol, and incubated at room temperature (28 ± 2^0C) for 5-7 days. The percentage of seeds infected with *Aspergillus* spp. was counted (this genus is the most frequently associated with egusi melon in Nigeria (Aboaba and Amasike, 1991; Bankole et al., 1999). For the germination tests, moist sand was sterilized by drying at 105^0C for 12 h, and filled into perforated pots. Three replicates of 100 seeds were put in the sand and incubated at 28 ± 2^0C. Germination was checked from 7-10 days. A seed was considered as germinated if normal seedlings emerged as described by AOSA (1981).

Sensory evaluation

The sensory evaluation of the organoleptic qualities of the stored seeds was carried out at the end of 6 months storage by ten semi-trained panelists (six females and four males), students of the Faculty of Science, Olabisi Onabanjo University. The panelists were asked to rate the seeds after the shells have been removed for flavour, texture, colour and general acceptability on a 9 point hedonic scale (Larmond et al., 1991) from 1 to 9, where 1 = extremely disliked to 9 = extremely liked.

Statistical analysis

Data in percentages were transformed to arcsine values, while the aflatoxin levels were log transformed, then data were subjected to analysis of variance using the SUPERANOVA (Abacus Concepts Inc CA, USA) computer and significant differences between means were determined by the least significant difference technique at 95% confidence level (Peterson, 1985).

RESULTS AND DISCUSSION

The oil content of uninoculated melon seeds was 53.8%, but this was significantly reduced by all the four fungi (Table 1). Highest reduction of 19.1% in oil content was effected by *A. flavus* in untreated seeds, followed by *A. tamarii* (15.8%) and the least reduction by *P. citrinum* (13.6%). Fungi consume the oil in invaded seeds (Chakrabarti, 1987), thus the decreased oil content observed in inoculated seeds. Several authors have reported *A. flavus* and *A. tamarii* to be highly lipolytic (Eggins, 1963; Chakrabarti, 1987). The oil content of seeds treated with 1.0 and 2.0% (w/w) of ground leaves was not significantly different from that of the untreated inoculated seeds. However, treatment of seeds with higher dosages (5.0 and 10.0%, w/w) of ground leaves resulted in significantly higher oil content than that of the untreated inoculated seeds. The major constituent of melon seeds is oil, thus its percentage and quality are very useful criteria for determining the extent of deterioration.

The free fatty acid content of the untreated seeds varied from 5.4% with *P. citrinum* inoculated seeds to 10.6% with *A. flavus* inoculated seeds compared to 0.4% in uninoculated seeds (Table 2). All the levels of application of ground leaves resulted in significantly lower free fatty acid content in treated seeds compared to the control. The free fatty acid content decreased as the

Table 2. Effect of dry ground leaves of *C. citratus* on the free fatty acid content of melon seeds inoculated with different fungi.

Fungus	Dosage (wt/wt)					
	0	**1.0**	**2.0**	**5.0**	**10.0**	**LSD**
A. flavus	10.6	6.5	6.3	3.6	1.4	2.1
A. niger	7.7	5.1	4.9	2.6	2.1	1.9
A. tamarii	10.3	6.8	5.9	2.5	1.6	2.4
P. citrinum	5.4	2.9	2.6	1.1	0.7	1.3
LSD	3.6	2.7	2.2	1.7	1.3	

Table 3. Effect of dry ground leaves of *C. citratus* on the peroxide values (meg/kg) of melon seeds inoculated with different fungi.

Fungus	Dosage (wt/wt)					
	0	**1.0**	**2.0**	**5.0**	**10.0**	**LSD**
A. flavus	75.3	68.3	52.7	33.4	10.5	20.6
A. niger	58.2	40.5	27.7	13.4	6.55	15.4
A. tamarii	71.5	60.2	53.8	37.4	16.7	18.2
P. citrinum	46.6	39.3	21.5	10.5	8.8	16.1
LSD	13.8	15.2	11.8	8.6	5.2	

Means of three determinations.

dosages of the ground leaves increased. The liberation of free fatty acid from the constituent glycerides leading to the development of rancidity is a major form of deterioration caused by associated fungi in oil seeds (Ogundero, 1981).

The peroxide value in uninoculated seeds was 3.3 meg/kg, but was significantly higher in fungi-inoculated seeds. The increase in peroxide values indicates the development of rancidity (Adebiyi et al., 2002). Treatment of melon seeds with dry ground leaves of *C. citratus* resulted in reduced peroxide values, but the value recorded in seeds treated with 1.0% (w/w) of powdered *C. citratus* was statistically comparable to that of inoculated untreated seeds. Thereafter, significantly lower peroxide values were obtained in seeds subjected to higher dosages of dry leaves.

Generally, the results indicate that the extent of mould deterioration varied with the dosage of plant products. However, the powdered product did not completely inactivate the fungi, as the deterioration parameters studied were still higher than those of the uninoculated seeds.

Table 4 shows that the oil content was significantly lower, while the free fatty acid and peroxide values were significantly higher on the untreated seeds inoculated with aflatoxigenic *A. flavus* strains than on treated-inoculated seeds. The oil content increased, while the free fatty acid and peroxide values decreased as the dosages of essential oil increased. The effect of dry leaves on the oil content, free fatty acid and peroxide values at 10% (w/w) was only comparable to that of lower levels of essential oil application (0.1%, 0.25%, v/w). There were no significant differences in the oil content of inoculated seeds treated with 0.5 and 1.0% (v/w) oil of *C. citratus* and the uninoculated seeds.

The present results show that the essential oil and powdered leaves of the medicinal plant *C. citratus* are able to alter the rate of deterioration in melon seeds inoculated with toxigenic *A. flavus*. The essential oil proved to be fungistatic at lower dosages (0.1% and 0.25%, v/w), while at higher concentrations (0.5% and 1.0%, v/w) it became fungitoxic. Similar results were obtained by Fiori et al. (2000) who found that the oil of *C. citratus* provided 100% inhibition of mycelia growth and germination of spores of *Didymella bryoniae*, causal agent of the gummy stem blight of melon crop. The essential oil was more toxic to the fungi than the powdered leaves despite the fact that lower concentration of the former was used. This probably indicates that the fungicidal effect of the dried plant is due to the high concentration of essential oil. In similar studies, extracts from *C. citratus* were shown to have fungicidal potential against 10 dermatophytes and *A. fumigatus* (Kishore et al., 1993). The oil of *C. citratus* has also been shown to possess antibacterial action against many bacteria including *Pseudomonas aeruginosa* (Cimanga et al., 2002).

The mean level of aflatoxin B_1 detected in melon seeds inoculated with toxigenic *A.flavus* was 65.3 ng/g after 14 days of incubation (Table 4). Aflatoxin production was significantly reduced with 0.1 and 0.25% (v/w) essential oil and 10% (w/w) of dry ground leaves. Aflatoxin was not

Table 4. Effect of essential oil (0.1-1.0%, v/w) and powdered leaves (10.0%, w/w) from *C. citratus* on some biochemical parameters and aflatoxin B_1 production in shelled melon seeds inoculated with toxigenic isolates of *Aspergilus flavus* and incubated for 14 days.

% Concentration (v/w grain)	Oil Content (%)	Free fatty content (%)	Peroxide values (meg/kg)	Aflatoxin B_1 (ng/g)
Control 1 (inoculated)	45.7	6.7	59.3	65.3
Control 2 (uninoculated)	53.8	0.37	3.5	nd
0.1	50.6	2.3	5.5	15.3
0.25	51.5	1.7	5.3	8.3
0.5	53.3	0.37	3.6	nd
1.0	53.7	0.35	3.7	nd
Ground leaves	50.4	2.7	5.1	17.3
LSD	4.6	2.2	7.3	12.5

Minimum detectable level of aflatoxin B_1 = 5 ng/g.
nd = aflatoxin B1 not detected.

Table 5. Incidence of diseased seeds in melon seeds treated with dry ground leaves, essential oil of *Cymbopogon citratus* and Iprodione during storage.

Treatments	Sampling period (months)							
	0	1	2	3	4	5	6	LSD
Oil (0.5%, v/w)	0	0	1.7a	2.5	4.3	5.7	7.3	3.5
Dry ground Leaves	0	0	3.3	5.5	8.0	12.3	18.6	5.2
Iprodione	0	0	0.7	2.3	5.0	6.5	9.3	3.2
Control	0	0	3.7	8.3	15.3	24.6	38.5	10.1
LSD	NS	NS	NS	2.5	3.4	6.2	8.1	

Table 6. Percentage incidence of *Aspergillus* spp in melon seeds treated with dry ground leaves, essential oil of *Cymbopogon citratus* and Iprodione during storage.

Treatments	Sampling period (months)							
	0	1	2	3	4	5	6	LSD
Oil (0.5%, v/w)	2.3	2.5	4.3	5.7	7.5	11.7	15.3	4.5
Dry ground Leaves	2.3	3.3	6.7	10.5	13.7	19.3	26.6	6.6
Iprodione	2.3	2.4	3.7	4.5	7.3	9.0	13.7	3.8
Control	2.3	4.0	7.3	14.0	20.3	32.3	42.7	9.7
LSD	NS	NS	NS	4.2	5.6	7.7	10.2	

NS = No significant difference.

detected in seeds treated with 0.5 and 1.0% (v/w) of essential oil. The aflatoxin level detected in seeds treated with dry ground leaves was statistically comparable to that of the lowest level of essential oil (0.1%, v/w). That aflatoxin was not detected in inoculated seed treated with higher dosages of oil of *C. citratus* further confirm its fungitoxic properties.

The treatment of seeds with the essential oil, powdered leaves and fungicide (iprodione) significantly reduced the proportion of diseased seeds compared to the untreated check over the 6 months storage (Table 5). The incidence of visibly diseased seeds with oil treatment was not significantly different from that of fungicide (iprodione) treatment, but the dry leaf treatment was significantly less effective. The incidence of diseased seeds has been found to be a good predictor of the extent of mould deterioration and aflatoxin contamination in melon seeds (Bankole et al., 1999; Bankole and Adebanjo, 2004) and often is a major parameter that determines the market price of melon seeds, since this is one of the obvious properties that buyers could easily identify.

The infestation level of *Aspergillus* spp did not significantly differ in the first two months in farmers stores' (Table 6). Thereafter, the infestation levels differ,

Table 7. Percentage germination of melon seeds treated with dry ground leaves, essential oil of *Cymbopogon citratus* and Iprodione during storage.

Treatments	Sampling period (months)							
	0	1	2	3	4	5	6	LSD
Oil (0.5%, v/w)	97.3	96.7	93.3	93.7	89.3	87.5	82.7	6.2
Dry ground Leaves	96.7	95.0	91.7	90.3	82.7	76.5	69.7	9.7
Iprodione (Untreated)	97.7	97.3	94.5	93.3	91.3	86.0	84.5	5.9
Control	97.3	94.0	90.3	83.3	77.0	65.7	47.3	10.3
LSD	NS	NS	NS	3.3	6.5	10.2	13,5	

NS = No significant difference.

Table 8. Effect of oil (1.0%, v/w) and powdered leaves (10.0%, w/w) from *C. citratus* and iprodione on some biochemical parameters and aflatoxin B_1 contamination in melon seeds after 6 months in farmers' stores.

Treatment	Oil Content (%)	Free fatty content (%)	Peroxide values (meg/kg)	Aflatoxin B_1 (ng/g)
Untreated seeds (before storage)	53.8	0.4	3.5	nd
Untreated seeds (after 6 months)	46.3	3.3	53.6	45
Essential oil (0.5%, v/w)	51.7	0.6	10.7	nd
Powdered leaves (10%, w/w)	50.2	0.9	22.3	nd
Iprodione (0.3%, w/w)	52.5	0.7	9.4	nd
LSD	3.6	2.3	8.4	

nd = not detected.

Table 9. Sensory evaluation of melon seed subjected to different treatments after 6 months in farmers' stores.

Treatment	Parameters			
	Flavour	Texture	Colour	Overall acceptability
Essential oil	6.8	7.1	7.5	7.3
Dry ground Leaves	6.5	6.4	6.9	6.9
Iprodione	7.2	7.3	7.3	7.4
Control (Untreated)	6.1	6.2	5.5	6.1
LSD	0.7	0.8	1.1	0.7

Data represent mean of ratings done by 10 panellists on a nine point scale where 1 = extremely disliked, 2 = very much disliked, 3 -= moderately disliked, 4 = slightly disliked, 5 = neither liked nor disliked, 6 = slightly liked, 7 = moderately liked, 8 = very much liked and 9 = extremely liked.

when the untreated seeds had the highest level of *Aspergillus* spp. Among the different treatments, the infestation level was significantly higher in seeds treated with dry ground leaves. There was no significant difference in the infestation levels between seeds treated with essential oil and fungicide (iprodione).

Table 7 shows that treatments with the essential oil and dry ground leaves had no adverse effect on the initial seed germination. Seed germination decreased progressively with time in the stores, but it did not differ significantly in the first two months, but from the third month, it was significantly different for the various treatments. Seed germination of 82.7% in seeds treated with essential oil after 6 months was statistically at par with the 84.5% germination levels in fungicide treated seeds.

That *Aspergillus* spp was recovered from melon seeds at the beginning of storage at low incidence agrees with earlier reports (Bankole et al., 1999). The decreased seed germination observed with time may have been due to the increased number of storage fungi particularly *Aspergillus* spp, which can kill the seed germ due to increased metabolic activities by fungi (Bankole, 1998).

Table 8 shows that the oil content significantly decreased while the free fatty acid and peroxide values significantly increased in the untreated melon seeds

during storage (Table 8). The essential oil and powdered leaves of *C. citratus* appreciably protect the quality of the stored seeds. There were no significant differences between the oil content, free fatty acid and peroxide values of seeds protected with essential oil and fungicide (iprodione) and that of seeds before storage. Though, the free fatty acid content and the peroxide values were higher in melon seeds protected with powdered leaves of *C. citratus* than those of seeds protected with essential oil, the values were still lower than the 1% (Kuku and Adeniji, 1976) and 42-47 meg/kg (Evranud, 1993) maximum limit respectively in good quality oil seeds. The mean level of aflatoxin detected in unprotected seeds after 6 months in stores was 45 ng/g, but all the treatments suppressed the production of aflatoxin.

Table 9 shows that the flavour, odour and colour rating by panellists were significantly higher in treated seeds than that of the untreated seeds after 6 months in stores. The overall acceptability of the seeds was very much influenced by the flavour, texture and colour. Melon seeds treated with essential oil and iprodione had significantly higher acceptability than seeds treated with powdered leaves.

In Nigeria, melon seeds are stored for 6 months or more for use as food or seeds. The protection afforded by the essential oil and the powdered leaves (at high dosages) of *C. citratus* makes them suitable substitutes for fungicides in preserving the quality of melon seeds in stores. The efficacy of essential oil and powdered leaves of *C. citratus* against insects of stored products has also been described (Gbolade and Adebayo, 1993; Adebayo and Gbolade, 1994). The oil and ground leaves of lemon grass are not likely to pose any health risk considering their ethno medical use. Souza et al. (1986) observed that an infusion prepared from leaves of *C. citratus* orally administered to adult rats at doses up to 20 times than the corresponding human dosage did not induce any toxic effect. The fungitoxic properties of the oil of *C. citratus* have been found to be thermostable and unaltered for 7 months (Mishra and Dubey, 1994).

ACKNOWLEDGEMENTS

This work was funded by the International Foundation for Science, Stockholm, Sweden, and the Organisation of Islamic Conference Standing Committee on Scientific and Technological Cooperation, Islamabad, Pakistan through a grant to Dr S.A. Bankole. We also thank Miss O. Ajayi for the collection and preparation of plant material.

REFERENCES

Aboaba OO, Amasike J (1991). Storage of melon seeds. Nigr. J. Bot. 4, 213 – 219.

Adebayo TA, Gbolade AA (1994). Protection of stored cowpea from *Callosobruchus maculatus* using plant products. Insect Sci. Applic. 15: 185-189.

Adebiyi AP, Adeyemi IA, Olorunda AO (2002). Effect of processing conditions and packaging material on the quality attributes of dry-roasted peanuts. J. Sci. Food Agric. 82: 1465-1471.

AOSA (1981). Association of Official Seed Analyst's rules for testing seeds. J. Seed Technol. 62: 1-26.

Bankole SA (1993). Moisture content, mould invasion and seed germinability of stored melon. Mycopathologia 122: 123 – 126

Bankole SA (1994). Changes in moisture content, fungal infection and kernel germinability of maize in storage. Int. J. Trop. Plant Dis. 12: 213-218.

Bankole SA (1997). Effect of essential oil from two Nigerian medicinal plants (*Azadirachta indica* and *Morinda lucida*) on growth and aflatoxin B_1 production in maize grain by a toxigenic *Aspergillus flavus*. Lett. Appl. Microbiol. 24:190-192.

Bankole SA (1998). Postharvest fungal deterioration of melon seeds and its control.Ph.D Thesis, University of Ibadan, Ibadan, Nigeria. 219 p.

Bankole SA, Adebanjo A (2003). Mycotoxin contamination of food in West Africa: Current situation and possibilities of controlling it. Afr. J. Biotechnol. 2(9): 254-263.

Bankole SA, Adebanjo A (2004). Fungal infection and aflatoxin contamination of melon seeds from the humid forest and Northern Guinea savanna of Nigeria. J. Basic Microbiol. In Press.

Bankole SA, Eseigbe DA, Enikuomehin OA (1996). Mycoflora and aflatoxin production in pigeon pea stored in jute sacks and iron bins. Mycopathologia 132:155-160

Bankole SA, Ikotun B, Ekpo EJA (1999). Fungal deterioration of melon seeds stored in jute sacks and polyethylene bags in Ago-Iwoye, South Western Nigeria. Mycopathologia 146: 135-146.

Chakrabarti DK (1987). A review of deterioration of oil seeds with special reference to India. Int. Biodeter. 23: 137-157.

Cimanga K, Kambu K, Tona L, Apers S, DeBruyne T, Hermans N, Totte J, Pieters L, Vlietinck AJ (2002). Correlation between chemical composition an antibacterial activity of essential oils of some aromatic medicinal plants growing in Democratic Republic of Congo. J. Ethnopharmacol. 79: 213-220.

Eggins HOW (1963). Isolation and culture of fungi causing deterioration in Nigerian palm oil. Mycopathol. Mycol. Applicata 22: 201-213.

Evranuz EO (1993). The effect of temperature and moisture content on lipid peroxidation during storage of unblanched salted roasted peanuts: shelf life studies for unblanched salted roasted peanuts. Int. J. Food Sci. 28: 193-199.

Fan LT, Lai FS, Wang RH (1976) Cereal grain handling systems. In: Advances in Cereal Science Technology, vol 1 (Pomeranz A., ed.) St Paul, MN.

Fiori ACG, Schwan-Estrada KRF, Stangarlin JR, Vida JB, Scapim CA, Cruz MES, Pascholati SF (2000). Antifungal activity of leaf extracts and essential oils of some medicinal plants against *Didymella bryoniae*. J. Phytopathol. 148: 483-487.

Gbolade AA, Adebayo TA (1993). Fumigant effects of some volatile oils on fecundity and adult emergence of *Callosobruchus maculatus* F. Insect Sci. Applic. 14: 631-636.

Gong, YY, Cardwell KK, Hounsa A, Eggal S, Turner P C, Hall AJ, Wild CP (2002). Dietary aflatoxin exposure and impaired growth in young children from Benin and Togo: a cross-sectional study. Br. Med. J. 325: 20-21.

IARC (1993). Monograph Volume 56. Some naturally occurring substances: food items and constituents, heterocyclic amines and mycotoxins. IARC, Lyon, France,

Kishore N, Mishra AK, Chunsouria JP (1993). Fungitoxicity of essential oils aginst drermatophytes. Mycoses 36: 211-215.

Kuku FO, Adeniji MO (1976). The effect of moulds on the quality of Nigerian palm kernels. Int. Biodeter. Bull. 12: 37-41.

Lalith K, Vdyasekaran P, Govindaswamy CV, Draiswamy S (1971). Reduction of oil content of castor seeds due to storage fungi. Cur. Sci. 40: 263-273.

Larmond E, Butter G, Mackie DA, Paste LM (1991). Laboratory methods for sensory evaluation of food. (Research Branch, Agriculture Canada Publication 1864/E). Ministry of Supply and Services, pp. 64-67.

Mishra AK, Dubey NK (1994). Evaluation of some essential oils for their toxicity against fungi causing deterioration of stored food

commodities. Appl. Environ. Microbiol. 60: 1101-1105.
Ogundero VW (1981). Isolation of thermophilic and thermotolerant fungi from stored groundnuts and determination of their lipolytic activity. Int. Biodeter. Bull. 17: 51-56.
Pearson (1970). The Chemical analysis of foods, 6th ed, London, Longman Group, pp. 535-537.
Peterson RG (1985). Design and analysis of experiments. Marcel Dekker, Inc. New York and Bassel , 429pp.
Oyolu O (1977). A qualitative and quantitative study of seed types in 'egusi' (*Colocynthis citrullus* L.). Trop. Sci. 19: 51-61.
Stack ME, Pohland AE (1975). Collaborative study of a method for chemical confirmation of the identity of aflatoxin. JAOAC 58: 114-116.
Souza FLM, Lodder HM, Gianotti FO, Ferreira TM, Carlini EA (1986). Pharmacology of lemon grass (*Cymbopogon citrates*) 11. Effect of daily two months oral aministration in male and female rats. J. Ethnopharmacol. 17: 65-74.
Turner PC, Moore SE, Hall AJ, Prentice AM, Wild CP (2003). Modification of immune function through exposure to dietary aflatoxin in Gambian children. Environ. Health Perspect.111: 217-220.