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International Digital Organization for Scientific Research ISSN: 2579-0730
IDOSR JOURNAL OF BIOLOGY, CHEMISTRY AND PHARMACY 3(1) 158-167, 2019.

Studies on the Nutritional Qualities of Fungal Infected Melon Seeds Sold in Eke Imoha Market, Onueke Ezza, Ebonyi State, Nigeria

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ABSTRACT

Melon is grown mainly for its seeds, which forms a major soup ingredient and a common component of daily meals in Nigeria and West African at large. Some are used as a favorite food seasoning, while others are roasted and grounded into a spread like peanut butter e.t.c. This research was carried out to analyze the nutritional qualities of fungal infected melon seeds sold in EkeImoha market, OnuekeEzza, Ebonyi State. The standard blotter method was used for the isolation of the fungi. *Rhizopusstolonifer, Aspergillusniger, Aspergillusflavus, Aspergillustereus, Penicilliumitalicum* and *Penicilliumfuniculosum* were the fungi associated with the contamination of melon seeds bought from Eke Imoha market in Onueke Ezza, Ebonyi State, Nigeria. The result of the analyses of the apparently healthy and contaminated melon seeds revealed that, there was a decrease in the nutritional value of melon seeds infected by fungi. The value of crude protein was higher in the healthy seeds than the infested seeds with the mean value of 36.82±0.17 and 30.32±0.06 respectively. A gradual increase was observed in the mean value of crude fibre (1.40±0.03) that was infected with fungi than the mean value of crude fibre (1.36±0.01) of the non-infested melon seeds. There was an increase in the mean value of the moisture content (5.78±0.01) in infected *Citrulluscolocynthis* seeds. There was also an increment in the mean value of *carbohydrate* (14.21 ± 0.03) in infected *Citrulluscolocynthis* seeds. Whereas, the fat content in the analyzed seeds was significantly higher (41.42±0.02) in the apparently healthy (41.42±0.02) *Citrulluscolocynthis* seeds than in the infected (39.34±0.04) *Citrulluscolocynthis*seeds. Conclusively, poor handling, storage period and high moisture contents predisposes the melon seeds to fungal contaminations.

Keywords*:* Melon seed, Onueke, Apparently Healthy, Infected, Proximate analyses.

INTRODUCTION

Melon plays vital roles in the farming system and in the well-being of West African rural farmers as a good source of energy, weed suppressants and for soil fertilization [1]; [2]. It is also used as mulch, leaving high residual nitrogen in the soil after harvesting. Melon is one of the most economically important vegetable crops worldwide and is grown in both temperate and tropical regions [3]. A high-energy, high-protein concentrate, melon seed ideally complement Africa's prevalent diets based on starch-rich grains (rice, sorghum and maize, for instance) and roots (notably cassava, yam and potato).

Melon is a vital tool against marasmus (lack of calories), kwashiorkor (lack of protein), and other debilitations [4]. Use of melon as local medicine is attributed to its biomedical properties and efficacy in the treatment of some ailments as reported by [5].

As a traditional food plant in Africa, this vegetable has potential to improve nutrition, boost food security, foster rural development and support sustainable land care (National Research Council,

2006). Melon has been recognized as an affordable source of vitamins and micronutrients especially in the rural areas. There is also a prospect for use of the melon seed in the improvement of infant nutrition in view of its high protein and fat content [6].

In general, the kernel of *Citrulluscolocynthis* contains about 50 percent oil, 30 percent protein, 10 percent carbohydrate, 4 percent ash, and 3 percent fiber [7].. *Citrulluscolocynthis* is an excellent source of arginine, methionine, and tryptophan [8]. The seed flour is rich in micronutrients (vitamins and minerals), and could therefore be used in food formulations especially in regions with low milk consumption such as West Africa [9]. In general, however, significant growth improvement was reported when *Citrulluscolocynthis* flour supplemented traditional West African diets [10].

Citrulluscolocynthis seeds contain about 53 % oil, 28 % protein (60 % in defatted meal), 11 % starch and soluble sugars [11]. *Citrulluscolocynthis* seeds contain a fairly high amount of unsaturated fatty acid and Linoleic acid, suggesting a possible hypocholesteronic effect [12]. The melon seeds are rich in fatty acids such as myristic, palmitic, stearic, oleic, linoleic and linolenic acid. It is reported that the de-oiled cake can be incorporated in the cattle feed of milking cows up to 25 % and it did not exhibit significant effect on the milk yield [13]. *Citrulluscolocynthis* seed oil is edible; its composition is similar to that of soybean oil. Refining and washing with citric acid removes its bitter taste [14]. The protein content of seeds of *colocynthis* was found to be 8.25 % and rich in lysine, leucine and sulfo-amino acids viz., methionine [15]. Melon (*colocynthis*) kernels contain oil (52 %), protein (28.4 %), fiber (2.7 %), ash (3.6 %) and carbohydrate (8.2 %) [16]. These are good sources of essential amino acids such as arginine, tryptophan and methionine and vitamins (B1, B2, and Niacin) and Minerals (Ca, Mg, Mn, K, P, Fe and Zn).

The content of essential amino acids in the proteins of the flour makes it a good vegetable protein ingredient [17]. The fiber in the 10 % hull flour contributes nutritionally to it. Significant growth improvement was reported when *C. colocynthis* flour supplemented traditional West African diets either alone or with other plant proteins [18]. Feeding studies were not performed in this study but an investigation conducted elsewhere indicated that the biological indices of protein quality for *C. colocynthis* were lower than values obtained for soybean [19]. The first and second limiting amino acids in the flour are lysine and threonine, respectively [20]. [21] noted that lysine and methionine were the first and second limiting amino acids, respectively of watermelon seeds, a close relative of melon. Histidine has been known to be an essential amino acid for infants [22] and the possibility that histidine is equally essential for a normal adult has also been suggested [23]. Thus the low content of histidine in *C. colocynthis* seed should be considered in the use of this product in food formulations, especially if the foods are intended for infants. *C. colocynthis* seed flour contains several micronutrients (vitamins and minerals) that could contribute significantly to the diet [24]. The potential for *C. colocynthis* seed flour as sources of calcium and niacin is encouraging to the low milk-consuming regions of lower West Africa where *C. colocynthis* cultivation thrives [25].

The family Cucurbitaceae, has been reported to have quite a number of medicinal uses ranging from antiviral, antidiabetic, antiulcerogenic, antioxidant and hepatoprotective [26] to antihelmintic [27], antimalarial [28], anticancer [29]; [30] to cardioprotective properties [31].

Also, *Cucumismelo* (musk melon or cantaloupe) has been shown to possess useful medicinal properties which include anti-oxidant, free-radical scavenging, anti-platelet, anti-ulcer, anti-microbial, anti-cancer, anti-diabetic, anti-helmintic, anti-

fertility, analgesic and anti-inflammatory [32].

*Citrulluscolocynthis*is one of the most potent fruits for managing diabetes mellitus for a few reasons [33]. There are significant levels of charntin, peptides that resemble insulin, and alkaloids within the fleshy fruit of *Citrulluscolocynthis* [34]. All of these components actively affect the levels of blood sugar, mainly in reducing it.

*Citrulluscolocynthis*is a source of many different antioxidants that make it a powerful defense mechanism against illness in the body [35]. Antioxidants scavenge the body for free radicals, dangerous compounds released during cell metabolism that can cause any number of different illnesses [36]. By adding *Citrulluscolocynthis* to their diet, it can greatly improve the chances of defending against very serious diseases, including heart attack, kidney damage, and liver failure [37].

[38] reported a 0.46 % w/w of linolenic acid present in egusi melon oil. This may suggest why it is used in some parts of eastern Nigeria as remedy for benign prostate hyperplasia and prostate cancer. [39] have documented successful inhibition of hormonally-induced benign prostate hyperplasia (BPH) in Wistar rats by the seeds of another member of the Curcubitaceae family, the fluted pumpkin (*Telfairiaoccidentalis* Hook f.).

The antifungal and antibacterial qualities of *Citrulluscolocynthis*make it ideal for fighting off various fungal infections, and also helping to rid the bloodstream of those toxins before they can do any more damage. Specifically, in terms of infections and skin health, *Citrulluscolocynthis*has been useful in treating ringworm and psoriasis [40]. Furthermore, its anti-inflammatory qualities reduce the irritating itching associated with such skin conditions and infections [41]. The juice extracted from the leaves can be the best cure for these conditions, topically applied to the affected areas [42]. [12] using animal models, observed that egusi melon oil has the ability to improve serum and liver lipid profiles and offer protection against resultant lipid peroxides from consumption of high fat diet, thereby conferring an improved antioxidant status. [11], in another animal studies reported that egusi melon was found to show inhibitory activity against lecithin.

[15] reported that socio-cultural uses of melon include household food, income generation, gift and seeds. Almost all the big markets in Nigeria, Benin, Cameroon, Ghana, Togo, and other nearby nations sell the seed. Melon is in high demand in tropical markets, especially in the peri-urban and urban markets [33]. It is also exported to Ethiopia and Sudan where the consumption is high and the extracted yellow oil is in high demand [3].

[27] reported that fungi are the major cause of spoilage of grains and seeds, and probably ranks second only to insects as spoilage organisms. Fungi of the genera *Aspergillus*and *Penicillium*are widely distributed storage fungi of *C. colocynthis* seeds, causing seed discolorations, decreased nutritive value, increases in free fatty acid and peroxide values, decreased seed germination, producing a number of toxic metabolites including aflatoxin [23]: [24].

[40] observed that fungi like *Aspergillus niger*, *Aspergillusflavus*, *Alternariadianthicola*, recorded discolouration, rotting, shrinking, seed necrosis, loss in germination capacity and toxification in oil seeds. [2] have reported *Aspergillus flavus, A. niger, Rhizopusstolonifer, Burgoanigra* and *Fusarium*sp. in stored *Citrulluscolocynthis* seeds. According to [8], seed deterioration constitutes a major constraint to all year round availability of *Citrulluscolocynthis* in Nigeria and other parts of the world.

[12] reported that fungal infection is one of the major setbacks on *Citrulluscolocynthis* production in South-Eastern Nigeria. Some of the fungal pathogens of *Citrulluscolocynthis* include *Sclerotiumrolfsii, Botryodiplodiatheobromae, Cercosporacitrulina,*

Alternariacucumerina, Collectotrichumlagenarium, Fusariumoxysporium and *Aspergillusspp* [9].

One major problem that besets *C. colocynthis* seeds is that they deteriorate quickly in storage due to fungal activities [26]. Aflatoxins have been associated with elevated rate of liver cancer, stunted growth and immunotoxicity in West Africa [14] [15].

The conditions of the stored product determine the extent of invasion of the stored product. The environmental factors that aid the development of fungi in stored products include moisture content [23], temperature and aeration [5], pH [6], relative humidity [7]. However, the effects of these storage fungi on stored products include deterioration and spoilage of stored products [34], reduction of market value [12] and production of chemical substances that are toxic to human health [27]. The preventive measures that can be employed for the growth of the storage fungi are biological control [11], chemical control [2] and physical control [9].

MATERIALS AND METHODS

Sample collection

Peeled melon seeds were bought from Eke market, Onueke Ezza in Ezza South of Ebonyi State and were transported to Department of Applied Biology laboratory, Ebonyi State University, Abakaliki for treatment and chemical analysis.

Isolation and identification of the Fungi

The standard blotter method of the International Seed Testing Association (I.S.T.A) (1976) was used for the isolation of the fungi. Three layers of Whitman filter paper were placed in the Petri dishes and autoclaved at 121oC for 15 minutes. After the autoclaving the filter papers were moistened. The melon seeds were soaked in 1 % hypochlorite for two minutes, and washed in three changes of distilled water and incubated at 25 ± 2°C for 7 days. Pure culture was prepared from the fungal colonies observed on the melon seeds.

Potato Dextrose Agar (PDA) was used for the pure culture preparation. The PDA was prepared by dissolving 39 gram into 1000 ml of distilled water and autoclaved at 121°C for 15 minutes. The medium was poured into autoclaved Petri-dishes and allowed to gel. The different fungal colonies were aseptically placed using inoculation loop. Identification of the fungi was carried out according to [17].

Proximate Analyses of the Nutrients of the apparently healthy and infected melon seeds.

The contaminated melon seeds were thoroughly washed and dried, and were referred to as infected seeds, while the un-contaminated melon seeds were referred to as apparently healthy seeds. The apparently healthy and contaminated seeds were analyzed in triplicate for carbohydrate, crude fibre, moisture, protein, lipid (fat) and ash using standard method of [40].

Determination of moisture content

A laboratory crucible was washed and dried in an oven at 105 ± 2 °C for 1 hour. It was then weighed after cooling in a desiccator. 10g each of the pulverized treated *Citrulluscolocynthis* seeds was then added to the crucible and the total weight determined. The crucible, together with its contents was transferred into an oven at 105±2°C and dried for 3 hours, after which it was weighed. This process was repeated until difference in weight between two successive dryings was less than 0.1g. The difference in weight between the original sample and the dried sample was calculated using the formula according to [3] [4] and modified by [5]:

% weight loss = % Moisture content =

$$\frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where
W1 = initial weight of the empty dish
W2 = weight of dish + undried sample
W3 = weight of dish + dried sample
% Dry matter = 100% - % Moisture content

Determination of Crude fibre

About 5g each of the pulverized treated *Citrulluscolocynthis* seed was put into 200ml of 1.25 % sulphuric acid and boiled gently for 30 minutes and filtered through a muslin cloth into a Buchner funnel. The residue obtained was washed with hot sterile distilled water to remove acids. The acid-free residue was put into 200ml of 1.25 % sodium hydroxide and boiled for 30 minutes, after which it was filtered and then washed thrice with petroleum ether. The resulting residue was then put in a crucible and dried to constant weight after which it was cooled in a dessicator and weighed. Then the crucible containing the residue was then subjected to ashing in a muffle furnace at 300±10°C for about 30 minutes and cooled in a desiccator after which it was reweighed [37]; [38] [39].
The % crude fibre was calculated using the formula:

$$\frac{W_2 - W_3}{W_1} \times 100$$

Where W1 = weight of sample used.
W2 = weight of crucible + sample before ashing
W3 = weight of crucible + ashed residue

Ash content determination

About 10g each of the pulverized treated *Citrulluscolocynthis* seeds was placed into a pre-weighed and dried crucible and heated for 3 hours at 100°C. Afterwards, the crucible was transferred into a muffle furnace and the temperature slowly increased from 200 - 450°C, until ashing was complete, as indicated by white colour of the sample. The sample was then carefully removed and cooled in a dessicator to room temperature and reweighed immediately. [11] [12].
% Ash content was calculated using the formula:

$$\% \text{ Ash} = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where W1 = weight of dish only
W2 = weight of dish plus sample
W3 = weight of dish plus ashed sample

Determination of percentage fat content

About 5g each of the pulverized treated *Citrulluscolocynthis* seed was loosely wrapped in a filter paper and put into a thimble which is fitted to a dried round-bottom flask of known weight, containing 120ml of Petroleum ether. The sample was heated via a heating mantle and allowed to reflux for 6 hours after which the thimbles were removed. The solvent was then recovered by evaporation and the extracting flask with its oil content was dried in an oven at 60°C for 30 minutes to remove any residual solvent. After cooling in desiccators, the flask was reweighed.
The difference in weight of the flask was received as mass of fat.
% Fat content was calculated using the formula [31] [32].

$$\% \text{ Fat} = \frac{W_2 - W_3}{W_1} \times 100$$

Where,
W1 = weight of initial empty extraction flask
W2 = weight of flask + extracted oil
W3 = weight of sample

Determination of protein content

Percentage crude protein was determined by Kjedhal method. The total nitrogen content was determined and multiplied by 6.25 to obtain the percentage crude protein. 0.5g each of the pulverized treated egwusi seed sample was mixed with 10ml of concentrated sulphuric acid in a Kjedhal digestion flask. One tablet of Kjedhal catalyst was added to it and the mixture heated under a fume cupboard until a clear solution was obtained in a

separate flask. The acid and other reagents were digested but without sample to form the blank control.
All the digests were carefully transferred to 100ml volumetric flask using distilled water and made up to the mark in the flask. A 100ml portion of each digest was mixed, distilled and the distillate collected into 10ml of 4% boric acid solution containing three drops of mixed indicators (bromocresol green and methyl red). A total of 50ml distillate was obtained and titrated against 0.02M sulphuric acid solution. Titration was done from the initial green colour to a deep red end point [36] [37].
The nitrogen content was calculated as shown below:

$$\%N_2 = \frac{100 \times N \times 14 \times V_f \times T}{W \times 1000 \times V_a}$$

Where,
W = weight of sample analyzed
N = Concentration of sulphuric acid titrant
Vf = Total volume of digest
Va = Volume of digest distilled
T = Titre value - Blank

Determination of carbohydrate content

The total carbohydrate was determined by subtracting the sum of percentage crude fat, percentage crude protein, percentage fiber and percentage ash from one hundred, thus: 100 -% crude oil + % crude protein + % fiber + % ash = % carbohydrate content.

RESULTS AND DISCUSSION

The results of this study showed that *Rhizopusstolonifer, Aspergillusniger, Aspergillusflavus, Aspergillustereus*, and *Penicilliumitalicum*and *Penicilliumfuniculosum* were the fungi associated with the contamination of melon seeds bought from Eke Imoha market in Onueke Ezza, Ebonyi State, Nigeria
.

Table 1: Proximate components of Healthy and Infected *C. colocynthis*seeds

Nutrient	Apparently Healthy seeds	Infected seeds
Crude protein	36.82 ± 0.17	30.32 ± 0.06
Crude fibre	1.36 ± 0.01	1.40 ± 0.03
Moisture	4.89 ± 0.04	5.78 ± 0.01
Ash	4.89 ± 0.21	3.14 ± 0.01
Carbohydrate	9.34 ± 0.04	14.21 ± 0.03
Fat	41.42 ± 0.02	39.34 ± 0.04

The result of the analyses of the apparently healthy and contaminated melon seeds revealed that, there was a decrease in the nutritional value of melon seeds infected by fungi. The value of crude protein was higher in the healthy seeds than the infested seeds with the mean value of 36.82±0.17 and 30.32±0.06 respectively. A gradual increase was observed in the mean value of crude fibre (1.40±0.03) that was infected with fungi than the mean value of crude fibre (1.36±0.01) of the non-infested melon seeds. There was an increase in the mean value of the moisture content (5.78±0.01) in infected *Citrulluscolocynthis* seeds. There was also an increment in the mean value of*carbohydrate* (14.21 ± 0.03) in infected *Citrulluscolocynthis* seeds. Whereas, the fat content in the analyzed seeds was significantly higher (41.42±0.02) in the apparently healthy (41.42±0.02) *Citrulluscolocynthis* seeds than in the infected (39.34±0.04) *Citrulluscolocynthis*seeds as shown in Table 1 above.
The isolated fungi from the melon seeds agreed with [6], who isolated *Aspergillus flavus, A. niger, Rhizopusstolonifer,*

Burgoanigra and *Fusarium*sp. from stored *Citrulluscolocynthis* seeds. These fungi are responsible for seed decay and hence a reduction in the supply of the melon seeds. [8] stated that seed deterioration constitutes a major constraint to all year round availability of *Citrulluscolocynthis* in Nigeria and other parts of the world. [23] reported that some of the fungal pathogens of *Citrulluscolocynthis* include: *Sclerotiumrolfsii, Botryodiplodiatheobromae, Cercosporacitrulina, Alternariacucumerina, Collectotrichumlagenarium, Fusariumoxysporium* and *Aspergillus spp.*

According to [27] fungi of the genera *Aspergillus*and *Penicillium* which attack the stored products are generally grouped into two categories namely field and storage fungi, which attack developing and matured seeds in the field and during storage respectively. Most of these fungal contaminations occur both in the field and after harvest and therefore adequate care needs to be implemented to prevent the contamination.

[36] [37] also reported that Fungi of the genera *Aspergillus*and *Penicillium*are widely distributed storage fungi of *C. colocynthis* seeds that cause seed discolorations, decreased nutritive value, increases in free fatty acid and peroxide values, decreased seed germination, and production of Mycotoxins.

The environment where the melon seed is stored also plays a major role in its contamination. This is supported by some researchers who stated that the environmental factors that aid the development of fungi in stored products include moisture content according to [5], temperature and aeration according to [8], pH according to [12] and relative humidity according to [26]. From this report it will be gathered that handling of melon is very difficult because of these factors that must be carefully managed before the contamination of the seeds is arrested.

There is enormous impact of fungal contaminations on melon seeds. According to some researchers, the impacts of these storage fungi on the stored products include deterioration and spoilage of stored products [26], reduction of market value [3] and production of chemical substances that are toxic to human health [18]. One major problem that besets *C. colocynthis* seeds is that they deteriorate quickly in storage due to fungal activities, [35].

The results of this finding was in line with the report of [39] who reported that deterioration caused by seed-borne microorganisms mainly fungi can result to deterioration in the proximate composition of cucurbits, comprising subsequent changes in organoleptic properties such as taste, flavour and texture.

The nutritional quality of melon(*C. colocynthis*) seeds was reduced in fungal contaminated *C. colocynthis* seeds when compared with healthy seeds in this study. This is supported by [40] who reported a decrease in the crude protein, crude fibre and total carbohydrate contents in seeds inoculated with fungal isolates for 14 days.

Fungi consume the oil in invaded seeds. The decreased oil content observed in deteriorated *Citrulluscolocynthis* seeds compared to healthy seeds is in line with the observation of [25]. Storage of seeds with their high moisture content promote mould invasion and affects the germinability of the seeds [31].

Some of the fungi produce mycotoxins, especially Aflatoxins, which predispose the consumers of the affected melon seeds to liver cancer, a food-borne carcinogenic agent [24]. [25] has shown that aflatoxins occur in detectable quantities in *C. colocynthis* found in Nigeria. Several outbreaks of mycotoxicosen disease in human and animals caused by various mycotoxins have been reported after the consumption of mycotoxin contaminated food and feed [36].

CONCLUSION

Fungi attack and devalue the fungal contaminated seeds sold in Eke Imoha market, Onueke. The handling and management of melon seeds is very difficult owing to the fact that the storage environment needs a lot of knowledge in handling the factors such as relative humidity, temperature and aeration and pH. The longer the storage period of the melon seeds, the contaminated they become.Therefore care must be taken to store melon seeds in a standard environment with control temperature and relative humidity.

REFERENCES

1. Aboloma, R.I. and Ogbunbusola, E.M. (2012a). Fungi associated with Irvingiagabanesis (Ogbono) and *Colocynthiscitrullus* (Egusi) seeds sold in markets in Ado-Ekiti, Ekiti State, *Nigeria Journal Natural Products Plant Resources,* **2**(3):423-426
2. Aboloma, R.I., Onifade, A.K. and Adetuyi, F.C. (2012b). Effect of deterioration on the proximate composition of some fruits of the family Curcurbitaceae. *Journal of Microbiology and Biotechnology Research,* **2**(1):240-243
3. Achigan-Dako**,** G.E., Fagbemissi, R., Ahanchade, A. and Avohou, H.T. (2006). Agronomic evaluation of threeEgusi species (*Cucurbitaceae*) used as food in Benin and development of a predictive model performance.*BiotechnologyAgron. Soc. Environ*, **10** (2): 121-129.
4. Achigan-Dako, G.E., Vodouche, S.R. and Sangare, A. (2008). Morphological characterization of local cultivars of *Lagenariesiceraria*(Cucurbitaceae) collected in Benin and Togo. *Belgium Journal of Botany*, **141** (1): 21-38.
5. Adeleke, E.E., Amadi, J.E. and Adebola, M.O. (2012). Studies on the Fungi Involved in the Deterioration of Stored Melon Seeds (*Citrulluscolocynthis* (L.) Schrad in Ilorin Metrpolisand Control. *Journal of Applied Sciences*, **15**(2): 10590-10602.
6. Aderiye, B.I. (2004). Contributory roles of microbes to human development. 10th Inaugural lecture, University of Ado Ekiti, Nigeria. Pp. 11 - 36.
7. Amusa, N.A., Kehinde, I.A. and Ashaye, O.A. (2002). Biodeterioration of the Africa Star Apple (*Artocarpuscommunis*) in storage and its effects on the nutrient composition. *African Journal of Biotechnology,***1**(2): 57 - 60.
8. A.O.A.C. (2005). Official methods of analysis (18th edition). Association of Official Analytical Chemists International, Maryland, USA.
9. Asoegwu. S.N. (1987). Tillage effects of egusi melon (*Colocynthiscitrillus*L.) production in Nigeria. *Proc. ofthe 12th Annual conference of Horticultural Society,* Pp 84-91.
10. Bankole, F. (1993). Profitability of Egusi Melon (*Citrulluslanatus*) production under sole and mixed cropping systems in Kogi State, Nigeria.*ARPN Journal of Agricultural and Biological Science,***3**(2):14-18.
11. Bankole, S.A., Osho, A., Joda, A.O. and Enikuomehin, O.A. (2005). Effect of Drying Method on the Quality and Storability of "egusi" Melon Seed (*Colocynthiscitrullus* L.), *African Journal of Biotechnology,***4**(8): 799-803.
12. Bankole, S.A., Ogunsanwo, B.M., Osho, A. and Adewuyi, G.O. (2005a). Fungal combination and aflatoxin B1 of egusi melon seeds in Nigeria. *Food Control,***17**(10):814-818

13. Bankole, S.A., Osho, A., Joda, A.O. and Enikuomehin, O.A. (2005a). Effect of drying method on the quality and storability of egusi melon seeds(*Colocynthiscitrullus* L.) *African Journal of Biotechnology,***4**(8):799-803
14. Bisognin, D.A. (2002). Origin and evolution of cultivated cucurbits. *Ciêcia Rural*, **32**: 715-723
15. Burell, N.J. (1974). Chilling in storage of cereal, grain and their products. *In*: C.M. Christensen (Ed.) American Association of cereal chemists. Inc., St. Paul MN.
16. Chiejina, N.V. (2006). Studies on Seed-borne Pathogens of Some Nigerian Melons, *Journal of Agriculture, Food, Environment and Extension,* **5**(1): 13-16.
17. Donli, N.D. and Gulaniin, A.E. (2001). Isolation and identification of fungal flora associated with melons in different storage facilities. *Small Wars Journal,***2**: 34-36
18. Ekundayo, C.A. and Idzi, E. (2005). Mycoflora and nutritional value of shelled melon seeds (*Citrullus vulgaris schrad*) in Nigeria. *Journal ofPlant Foods for Human Nutrition,***40**: 31 – 40.
19. FAO, (2002). *Bacillus cereus* and other *Bacillus* sp. U.S. Food and Drug Administration center for food safety and applied nutrition, Bed Bug Book. Food borne pathogenic micro-organisms and natural toxin handbook. http// www.cfsan.fda.gov/mow/chap12.htm
20. Giwa, S., Abdullah, L.C. and Adam, N.M. (2010). Investigating egusi (*Citrulluscolocynthis*L.) seed oil as potential biodiesel feedstock. *Energies,***3**:607-618
21. Gong, P., Chaluvaraju, S.A., Deepak, K.N., Amruthesh, A. and Shekar, S.H. (2002). Location and transmission of downy mildew pathogen *Plasmosparahalsteo* in sunflower seeds. *Seed Research,***32**: 108-110.
22. Gurudeeban, S. Satyavani, K. and Ramanathan, T. (2010). Bitter Apple (*Citrulluscolocynthis*): An Overview ofChemical Composition and Biomedical Potentials. *Asian Journal of Plant Sciences,* **9**: 394-401.
23. Hegazy, E.M. (2011). Effect of powder and essential oil of lemon grasss on aflatoxin production in dried water melon seed.*Life Science Journal,***8**(3):516-521
24. Kakde, R.B. and Chavan, A.M. (2011). Extracellular lipase enzyme production by seed-borne fungi under the influenc of physical factors. *International Journal of Biology,***3**: 95-100.
25. Khatri, L.M., Nasir, R., Saleem, M.K. and Valhari, M.U. (1993). Characteristics and chemical composition of *Citrulluscolocynthis. Pakistan Journal of Science Research,***36**: 384-384.
26. Kopple, J.D. and Swendseid, M.E. (1974). Nitrogen balance and plasma amino acid levels in uremie patients fed on essential amino acid diet. *American Journal of Clinical Nutrition*, **27**: 806-812.
27. Kuku, F.O. (1979). Deterioration of melon seeds during storage at various reltivehumidities. *Republic of Nigerian Stored Products Research Institute Technical Report,***7**: 65 - 67.
28. Makinde, E.A., Ayoola, A.T. and Akande, M.O. (2007). Effects of organo-mineral fertilizer application on the growth and yield of egusimelon.*Australian Journal of Basic and Applied Sciences,***1**(1):15-19
29. Mohanna, C. and Sharma, J.K. (1991). Seed pathology of forest tree species in India – present status practical problems and future prospects. *Commonwealth Forestry Research,***70**: 133-151.

30. Muller, J.D. (1991). Fungi and mycotoxin in stored products. In: B.R. Champ, Ettighealy, A.D. Hocking and J.J. Pitt (Eds.).ACIAR proceedings. Australian center for International Research. Pp. 126 - 135.
31. National Research Council (NRC), (2006). "*Egusi*". *Lost Crops of Africa: Volume II: Vegetables.* NationalAcademies Press. ISBN 978-0-309-10333-6.
32. Oyenuga, V.A. and Fetuga, B.L. (1975). Some aspects of the biochemistry and nutritive value of the watermelon seed (*Citrullusvulgaris*schrad). *Journal of Science, Food and Agriculture*, **26**: 843-846.
33. Ramakrishna, G., Azeemoddin, G. and Lakshminarayana, T. (1993). Processing of tumba seeds and oil. *Journal of Oil Technology Association,***25**: 3-5.
34. Rice, R.G., Graham, D.M. and Lowe, M.T. (2002). Recent ozone applications in food processing and sanitation. *Food Safety Magazine,***8**(5): 10 - 17.
35. Richard, J.L. and Wallace, H.A. (2001). Mycotoxins, National center for Agricultural utilization research, USDA/ARS.
36. Sanchez, A., Fuller, A.B., Yahiku, P.Y. and Baldsin, M.V. (1972). Supplementary value of blackeyed peas, peanuts and egusi seed on the typical West African diet of plant origin. *Nutr. Rep. Int.*, **6**: 171-179.
37. Sayed, M.D., Afifi, M.S. and Hassan, M.A. (1979). Investigation of the flavonoid content of certain vicia species growing in Egypt. *Bulletin of Faculty of Pharmacy, Cairo University,* **23**: 2-2.
38. Schippers, R. R. (2000). *African Indigenous Vegetables. An overview of the cultivated species.* Chatman, UK:Natural resources Institute/ACP-EU Technical Centre for Agricultural and Rural Cooperation.
39. Shaheen, A.M. and Hamed, A.I. (2003). Comparative studies and nutritional values of some weedy species collected from newly reclaimed areas (Western shore of Lake Nasser, Aswan, Egypt). *Egypt. Journal of Biotechnology,***13**: 176-186.
40. Theng, O.F.A. and Scrimshaw, B.N. (1968). Post-harvest seed-borne diseases associated with the seeds of three varieties of melonss, (*Arachishypogaea* L) Nwakara, Kaki and Campalla. *Agric. Biol. J. N. Am.,* **2:** 598-602.
41. van der Vossen, H.A.M., Denton, O.A. and El-Tahir, I.M. (2004). *Citrilluslanatus. In*: Grubben, G.J.H andDenton, O.A. *Plant resources of Tropical Africa 2 Vegetables.* Wageningen. The Netherlands; CTA,Leiden, the Netherlands: Backhuys Publishers, pp 185-191.
42. Yu, J., Chang, K.C., Ehrlich, J.W., Cary, P.K. and Bhatnagar, D. (2004). Clustered pathway genes in aflatoxin biosynthesis. *Applied Environ. Microbiol.*,**70**: 1253-1262.