International Journal of Agronomy and Agricultural Research (IJAAR)
ISSN: 2223-7054 (Print)
Vol. 2, No. 2, p. 1-7, 2012
http://www.innspub.net

RESEARCH PAPER OPEN ACCESS

Nutritive value and mycoflora of sun dried cocoyam chips during storage

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Received: 19 December 2011
Revised: 18 January 2012
Accepted: 19 January 2012

Key words: Mycoflora, cocoyam, proximate, nutritive value.

Abstract

The nutritive value and the mycoflora of sundried cocoyam chips were investigated during six months storage. Seven fungi namely *Aspergillus flavus*, *Aspergillus niger*, *Aspergillus glaucus*, *Absidia corymbifera*, *Mucor* spp., *Rhizopus* spp., *Candida* spp were isolated during the study. The fungal count was found to increase as the storage time increased. The result of the proximate analysis (in %) showed that crude protein increased from 9.32 in freshly prepared samples to 11.36 in sundried samples stored for twenty four weeks. Also, carbohydrate content increased from 68.32 in freshly prepared samples to 69.52 in stored samples. Ash content increased from 3.55 to 4.20. Fat content decreased from 1.76 to 1.40 and the moisture content also decreased from 14.07 to 12.47 within the six months of storage. The result of mineral analysis (in mg/100g) showed that magnesium increased from 13.86 to 19.43, sodium increased from 15.53 to 16.55 but calcium was decreased from 23.50 to 23.21, zinc from 0.73 to 0.68 and phosphorus from 38.55 to 33.85. Copper, manganese and lead were not detected. This study has revealed the increase in the nutritional value and proximate analysis of sun dried cocoyam chips.

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Introduction

Cocoyam is a staple root crop that belongs to the *Araceae* family. The plant grows best in well drained light texture soils but can still tolerate heavy texture soil with pH from 5.5 to 6.5 (Raemaeker, 2001). It is an herbaceous monocotyledon plant of 1m height. The above ground stem consist of heart shaped leaves supported by soiled and long erect petioles. The underground corm is a compact structure filled with nutrients (Onokpise *et al.* 1999).

Cocoyam is considered as a food crop; corms, cormels are boiled, roasted or baked, leaves are rich in vitamins and minerals and are prepared as vegetable in West and Central Africa (Sandifola, 2002). It is an important crop in tropical and sub tropical areas because it provides carbohydrates, proteins, fats and vitamins (Tambong *et al.* 1997; Torres *et al.* 1994). It is highly nutritious with a considerable amount of carbohydrate (88.04%), ash (3.20%), crude protein (5.19%), crude fibre (2.86) and 0.40% ether extract (Aderolu, 2010). However, storage conditions have effect on the proximate and nutritive value of the stored root crops because of the growth of some spoilage fungi that strife in such conditions (Abaka and Norman, 2000).

The fungi that invade stored product are generally grouped into two categories namely field fungi which attack developing and matured seeds in the field and storage fungi which are predominantly species of *Aspergillus* and *Penicillium* which attack the stored products (Fagbohun *et al.* 2010).

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 (Amusa *et al.,* 2002), temperature (Abaka and Norman, 2000), aeration (Burell, 1974), pH (Aderiye, 2004), relative humidity (Fagbohun and Lawal, 2011). However, the effect of this storage fungi on stored products include deterioration and spoilage of stored products (Abaka and Norman, 2000; Ekundayo and Idzi, 2005), reduction of market value (Fagbohun *et al.*, 2010) and production of chemical substances that are toxic (Richard and Wallace, 2001). The preventive measures that can be employed for the growth of the storage fungi are biological control (Aderiye, 2004), chemical control and physical control (Rice, 2002).

However, the aims and objectives of this study were to study the nutritive value and the mycoflora of sun dried cocoyam chips during storage.

Materials and methods

Collection of Samples

The samples of fresh cocoyam were collected from Irona market, Ado Ekiti, Ekiti state, Nigeria. The cocoyam was peeled and immersed into warm water for ten minutes and sun dried for two weeks. The sun dried samples were stored in an air tight container insect container at 28^{o}C for six months. The samples were examined for the changes in the mycoflora and nutrients composition after each month of storage.

Isolation of fungi from the stored sun dried melon

Direct Plating: Samples of the sun dried cocoyam chips were examined randomly for external mouldness. Using a sterile dissecting forceps, the surface of the stored dried cocoyam chips were scrapped and was plated aseptically on Potato Dextrose Agar (PDA) plate and incubated at 28^{o}C for 5 - 7 days as described by Amusa (2001) and Arotupin and Akinyosoye (2001). The fungi cultures were subcultured until pure colonies were obtained by successive hypha tip transfer (Fagbohun and Lawal, 2011). The cultures were examined under the microscope for fruiting bodies, hyphae to determine the common fungi present.

Dilution Plate Method: This method was used to determine the type of fungi present in the stored sun dried cocoyam chips. About 1g of the sample grinded with 10ml of sterile distilled water. This was shaken thoroughly and 1ml of suspension was

pipetted into a sterile test tube containing 9ml of distilled water. This was thoroughly mixed together. The sample was serially diluted and 1ml each of aliquots of 10^{-5} and 10^{-6} were added to molten PDA plates. The plates were swirled gently to obtain thorough mixing and were allowed to solidify and incubated at 28°C for 5 - 7 days. The fungal colonies were counted every 24 h. Successive hyphae tip were transferred until pure cultures of each of fungus was obtained.

Washing Method: This was carried out by weighing 1g of the sundried cocoyam chips into 10ml of sterile distilled water in a beaker. This was shaken thoroughly and drops of suspension of contaminated water were introduced into petri dishes containing Potato Dextrose Agar. This was evenly spread on the agar plate with aid of a sterile glass spreader. The plates were incubated 28°C for 5 - 7 days and were observe for visible fungi growth.

Identification of mycoflora: The associated fungi were identified by their cultural and morphological features (Burnett, 1975; Alexopoulous et al. 1996; Dungan, 2006). The isolates were examined under bright daylight for the colour of the culture and further examination was carried out.

Needle mount preparation method: The method was carried out according to Tuite (1961), Crowley (1969), Burnett, (1975) and Dungan (2006) whereby fragments of the sporing surface of the initial culture was taken midway or between the centre and the edge of the colony. This was teased out in drop of alcohol on a sterilized glass slide using a botany needle. The fragments were stained by adding a drop of lactophenol blue. A cover slip was applied and the preparation was examined under X10 and X40 objective lens of the microscope.

Slide culture technique: From a plate approximately 2mm deep, 1cm² PDA was cut and placed on a sterile glass slide. Fungus was inoculated into the four vertical sides using a sterile needle. A sterile cover slip was placed on it so that it over lapped the medium on all sides. The preparation was placed on a suitable support in a petri dish containing blotting paper soaked in 20% glycerol in water. The preparation was kept moist at 28°C until adequate growth was observed. After removing the medium with scalpel, the fungus adhering to both cover slip and slide was examined (Crowley *et al.* 1969). A drop of alcohol was added followed by a drop of lactophenol blue and the preparation was covered and examined under the low power objective of microscope.

Proximate analysis: The proximate analysis of the samples for moisture, ash, fibre and fat were done by the method of AOAC (2005). The nitrogen was determined by micro-Kjeldahl method as described by Pearson (2002) the percentage Nitrogen was converted to crude protein by multiplying 6.25. All determinations were performed in triplicates.

Mineral analysis: The minerals of the samples were analyzed using the solution obtained by dry ashing the sample at 550°C and dissolving it in HCl (25ml) and 5% Lanthanum chloride (2ml), boiling, filtering and making up to standard volume with deionized water. Mn, Cu, Co, Zn, Fe, Mg, Na, and Ca were determined with a Buck Atomic Absorption Spectrometer (Buck Scientific, Model 200A/200, Inc. East Norwalk, Connecticut, U.S.A). Sodium was measured with a Corning 405 flame photometer (Corning Halstead, Essex, UK, Model 405) (AOAC, 2005). The detection limits had previously been determine using the methods of Techtron (1975) as Mn 0.01, Cu 0.005, Co 0.05, Zn 0.005, Fe 0.02, Mg 0.002, Ca 0.04, Na 0.001, ppm (all for aqueous solutions).

The optimum analytical range was 0.5 to 10 absorbance units with coefficient of variation of 0.05 to 0.40% phosphovanado-molybdate method using a Spectronic 20 colorimeter (Galenkamp, London, UK) (AOAC, 2005). All chemicals were BDH analytical grade.

Results and discussion

A total of seven fungi were isolated from stored sundried cocoyam chips based on their cultural and morphological characteristics. The fungi include *Aspergillus niger, Aspergillus flavus, Rhizopus* spp., *Candida* spp., *Aspergillus glaucus*, *Mucor* spp. and *Absidia corymbifera*. The summary of the fungi isolated from stored sundried cocoyam chips using various methods are shown on Table 1. In addition, results of the proximate and mineral analysis are shown on Tables 2 and 3 respectively.

Table 1. A summary of fungi isolated from stored sun dried cocoyam chips using various methods.

Weeks of storage	Fungi isolated
freshly prepared	A, B
4 weeks	A, B, E
8weeks	A, B, E
12 weeks	A, B, C, E, F
16 weeks	A, B, C, E, F, G
20 weeks	A, B, C, D, E, F, G
24 weeks	A, B, C, D, E, F, G

A = *Aspergillus niger*, B = *Aspergillus flavus*, C = *Rhizopus* spp., D = *Candida* spp., E = *Aspergillus glaucus*, F = *Mucor* spp., G = *Absidia corymbifera*

The results showed that *Aspergillus niger, Aspergillus flavus, Rhizopus* spp., *Candida* spp., *Aspergillus glaucus*, *Mucor* spp. and *Absidia corymbifera* were found to be associated with the stored sun dried cocoyam chips most of which are known to be surface contaminants of most agricultural products. They cause decay of agricultural produce thereby reducing their market and nutritional value (Amusa *et al.,* 2002). The fungi isolated using washing method are those capable of growing on the cocoyam chips. The fungi isolated by any of the three methods used could therefore be field or storage fungi (Ogundana, 1990).

In this study, there was an increase in the number of fungi isolated as the study progressed. Three fungi were isolated within the eight weeks of the study while five fungi were isolated after twelve weeks of the study. The number of fungi isolated increased to seven after the twentieth week of the study and remained through twenty fourth week. This result is in agreement with the findings of Okungbowa and Osagie (2009) who reported the progressive isolation of *Botryodiplodia theobromae, Rhizopus stolonifer, Mucor mucedo, Aspergillus niger, A. fumigatus, A. flavus* and *Penicillium digitatum A. ochraceus, Curvularia* spp., *Neurospora sitophila* from sun dried sweet potato stored for six months.

Moreover, Fagbohun *et al.* (2010), Fagbohun and Lawal (2011) also reported the progressive increase in species of fungi in sundried plantain chips and melon seeds respectively stored for twenty weeks. This progressive increase in the number of fungi species isolated may be due to the ability of the organisms to secrete extracellular enzymes capable of degrading the nutrient for the active growth of other organisms (Abiodun *et al.* 2007).

The proximate analysis (in %) of stored sundried cocoyam chips are shown on Table 2. It was revealed that the freshly prepared cocoyam chips had ash content (5.13), moisture content (16.32), crude protein (7.21), fat (3.20), crude fibre (1.48) and carbohydrate (88.40). However, after twenty four weeks of storage the % ash, moisture content, fat, crude fibre and carbohydrate decreased to 4.27, 12.47, 1.27, 1.40 and 69.56 respectively. This result is in agreement with the findings of Fagbohun *et al.* (2010) who reported the decrease in the % of moisture content, crude fibre, and carbohydrate of sundried plantain chips during storage. In contrast, % crude protein increased to 11.06. This result is in agreement with the findings of Koukou *et al.* (2010) who reported the percentage increase of crude protein of yam tubers stored for twenty four weeks. The decrease in the proximate may be due to fungal activity that caused changes during storage of the product. Nutrients are lost because of changes in

CHO, protein, lipids and vitamins (Abaka and Norman, 2000).

Table 2. A summary of the results of proximate analysis of sun dried cocoyam chips during storage (%).

Weeks of storage	Ash	MC	CP	Fat	Fibre	CHO
Freshly prepared	5.13	16.32	7.21	3.20	1.48	88.04
4 weeks	3.55	14.07	9.32	2.99	1.76	68.32
8 weeks	4.23	14.33	9.27	2.94	1.70	67.56
12 weeks	4.34	9.87	10.30	2.55	1.54	71.42
16 weeks	4.99	10.13	10.49	1.53	1.49	71.42
20 weeks	5.24	11.34	10.72	1.35	1.49	69.88
24 weeks	4.27	12.47	11.06	1.27	1.40	69.56

MC = Moisture Content, CP = Crude Protein, CHO = Carbohydrate

Table 3. A summary of the results of the mineral analysis of sun dried water melon seed during storage (mg/100g).

Storage in weeks	Na	K	Ca	Mg	Zn	Fe	P	Cu	Pb	Mn
Freshly prepared	14.42	35.01	20.23	12.22	17.33	0.70	37.42	ND	ND	ND
4	15.53	35.23	23.50	13.86	18.68	0.73	38.55	ND	ND	ND
8	16.25	35.84	26.21	19.21	22.16	0.93	39.48	ND	ND	ND
12	16.79	38.04	28.18	24.97	27.80	0.90	39.54	ND	ND	ND
16	17.53	41.48	36.64	18.76	23.69	0.88	36.85	ND	ND	ND
20	16.62	39.34	36.20	18.14	19.02	0.79	35.33	ND	ND	ND
24	16.55	36.54	23.21	19.43	14.96	0.65	33.85	ND	ND	ND

The mineral analysis of the sundried cocoyam chips during storage (in mg/100mg) are shown on Table 3. It was found that Na (14.42), K (35.01), Ca (20.23), Mg (12.22) in the freshly prepared cocoyam chips increased to 16.55, 36.54, 23.21, 19.43 respectively at twenty fourth week of storage. This is in agreement with the findings of Fagbohun *et al.* (2010) who reported an increase in the minerals of sundried plantain chips stored for twenty weeks. However, Zn (17.33), P (37.42) Fe (0.70) in freshly prepared cocoyam chips decreased to 14.96, 0.65, 33.85 respectively while Cu, Pb and Mn were not detected. This is in agreement with the findings of Echendu *et al.* (2009) and Alinnor and Akalezi (2010) who reported the decrease in Zn, P, Fe of cocoyam and white yam respectively stored for six months.

Conclusion

Cocoyam are of great economic importance and in order to maintain the quality, they should be stored under controlled environment that would not be favourable for the growth of fungal flora, thereby preventing deterioration of the stored cocoyam chips and reduction of the nutrients. This present study has revealed the various fungi associated with stored cocoyam chips. However, apart from good hygiene, proper handling and processing practice should be employed to reduce the contamination of stored cocoyam chips. The isolated fungi can degrade the cocoyam chip as substrate thereby reducing the market value, also making consumers especially the immunocompromised individual vulnerable to microbial infection.

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