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Assessment of microorganisms associated with deterioration of *Chrysophyllum albidum* fruits vended in Minna, Nigeria.

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ABSTRACT:Fifty samples of *Chrysophylum albidum* fruits (ripe, unripe and spoilt) were purchased from the central market, Minna, Niger state, Nigeria and analyzed for the presence of spoilage microorganisms, The results revealed the presence of the following organisms which includes *Aspergillus fumingatus*, *Aspergillus niger*, *Geotrichum* sp, *Candida albicans*, *Candida tropicalis*, *Cephalosporium* sp, *Mucor* sp, *Torulopsis candida*, *Trichophyton cutaneum*, *Penicillum notatum*, *Cryptococcus neoformans*; *Rhodotorula rubra, Candida quilliermondii*, and *Candida pseudotropicalis* among the fungi, while the bacteria isolate include and *Bacillus cereus*, *Bacillus subtilis*, *Streptococcus faecalis*, *Streptococcus neofoma* and *Staphylococcus aureus*. *Aspergillus fumigatus* was the most frequently encountered fungus with occurrence rate of 23.33%. *Cryptococcus neoformans*, *Rhodotorula rubra*, *Candida quilleirmondi* and *Candida pseudotropicalis* had occurrence of 3.3% respectively. *Bacillus cereus* was the most frequent followed by *Staphylococcus aureus* among the bacteria isolated. Microbial spoilage significantly reduced the proximate nutrient contents of the fruits when compared to healthy samples for instance: the proximate analysis of ripe fruits revealed the following: moisture 42.2%, crude protein 8%, carbohydrate 52.7%, ash 26.2% and lipid 13% while that of spoilt fruit revealed a reduction in its composition: protein 1.25% ash 19.6, lipid 5.0% moisture 30.2% except carbohydrate with 74.1%. The presence of these fungi decreased the nutritional composition of the fruits. The fruits must be sorted out during harvesting and more effort should be made to eliminate these pathogens.

Key words: *Chrysophyllum albidum,* deterioration, fruits

Introduction

The African star apple (*Chrysophylum albidum*) belong to the family Saponaceous has in recent times become a crop of commercial value in Nigeria (Bada, 1997and Umeh *et al*;2002). The fleshy pulp of the fruit is eaten especially as snack and relished by both young and old (Cenrad, 1999). The fruiting period is usually between December to April depending on the variety and the environmental condition.(Okafor, 2001). Okigbo (1975) reported that the fruit contains 8.8% protein, 17.1% oil, 20.9% sugar, 11.0% starch and several minerals.

The African star apple has been found to have highest content of ascorbic acid, 100-3,330mg of ascorbic acid per 100gm of edible fruit or about 1000times that of orange and 10times that of guava or cashew (Asenjo, 1988). Asenjo (1988) also reported that it is an excellent source of vitamin, irons, and flavour to diet.

However the damage caused by insect pests and pathogens is one of the major problems faced by fruits traders in many parts of Nigeria The star apple fruits is susceptible to attack by various pests and diseases leading to low germination, seedling mortality and reduction in quality and quantity of fruit yield (Adelaja, 1997). Naumova (1972) opined that good quality fruit is essential for high stable yield, provided other necessary conditions are optimal. However, production of high quality seeds is still not enough to maintain longevity unless seeds are protected from possible damage usually occasioned by abiotic factors during the storage period.

The fungi that infest fruits and seed are of two groups namely field fungi and storage fungi (Christensen and Ichaufman, 1979). Field fungi are those fungi that infest fruit and seeds as they are developing in the field or after these seeds are matured but before they are harvested. The common genera field fungi are *Alternaria*, *Cladosporum*, *Fusarium* and *Helminthosporum* species. These fungi may discolour fruits, cause death of the ovule, shriveling of the seed and weakening or death of embryo. The damage caused by

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field fungi is done at the time fruits are harvested and kept in the store (Shetty, 1992).The storage fungi are those that proliferate fruits in storage. One of the characteristics that they share in general is the ability to proliferate without moisture. They comprise several species of *Aspergillus* and a few species of *Penicillium*. All these have the ability to grow on grains and seeds whose moisture is in equilibrium with the relative humidity of 70-90% (Shetty, 1992, Oyeleke *et al.*, 2002).

Amusa *et al* (2003) isolated the following fungi from *Chrysophyllum albidum: Botrydiplodia theobromae, Rhizopus stolonifer, Aspergillus niger, Aspergillus tamarri, Aspergillus flavus, Fusarium* species, *Penicillium* species and *Trichoderma* species. All these fungal isolates except *Trichoderma* sp were to be pathogenic to the African star apple fruit. Neegard (1977) and Oladiran *et al* (2002) reported that storage fungi are known to cause reduction germinability and affect several other physiological changes in the seed/ fruits. The aims of this study are to isolate and identify the fungi responsible for the spoilage of *Chrysophyllum albidum*, to identify the effects of these fungi on the nutritive content of the fruit and suggest possible ways of controlling/eliminating such microbial spoilage by these pathogenic fungi.

Material and Methods

Fruits Samples

Fifty fruits of *Chrysophyllum albidum* were bought from the central market in Minna, Niger State, Nigeria. The samples bought ranged from ripe, unripe to spoilt. These samples were immediately transferred to the Microbiology Laboratory of the Federal University of Technology, Minna, Niger State.

Preparation of Media

The following media were prepared as described by Fawole and Oso (1988): Saboraud dextrose agar (SDA), Nutrient broth (NB), and Nutrient agar (NA).

Innoculation

Fruits samples were surfaced sterilized with chlorox and inoculated on Nutrient agar (NA) and Saboraud dextrose agar (SDA) plates respectively. The plates were incubated at 37°C for 24hours for plates of Nutrient agar and 28°C for 72hrs for plates of SDA. The stalks of the fruits were removed and serial dilution of the swab surface was done and 1ml each of the 10^{-5} was dispensed into petri dish labeled "ST" and molten agar of SDA and NA was poured into the plate respectively. The plate were swirled to allow proper mixing of the sample with the agar and then incubated as earlier described. The fruits were bisected with a sterilized blade and the inner surface was swab with a sterile swab and mixed with 1ml of sterile water. A serial dilution was done for all the samples. 1ml of the 10^{-5} was dispensed into the petri-dish labeled "I" and molten agar of SDA an NA was poured to the plates. These were rocked to allow proper mixing of the sample with the agar. These were later incubated at 37°C for 24hrs for NA and 28°C for 72hrs for SDA. Thereafter bacteria colonies were counted and characterized biochemically according to Fawole and Oso (1988).

Proximate Analysis of Chrysophylum albidum

Determination of ash

The ash content *of Chrysophylum albidum* was determined by dry-weight method as described by Peason (1975).

Determination of lipid

The lipid content was examined by method described by Peason (1975).

Determination of Crude Protein.

Crude protein content was determined by macro-Kjeldahl methods as descrbed by Peason (1975).

Determination of Carbohydrates

The total protein, Ash, lipid and fiber content will be subtracted from 100%, the result account for the carbohydrates.

Moistures Determination

The moisture content was determined by dry weight method as described by Peason (1975).

Results

Table 1 reveals that the spoilt fruit has the greatest contamination followed by ripe, and unripe fruit is relatively low.

Table 1: Total Microbial Load of *Chrysophylum albidum (cfu/ml)*

SAMPLE	*BACTERIA LOAD*	*FUNGAL LOAD*
RIPE FRUIT	1.4×10^6	1.0×10^5
UNRIPE FRUIT	1.2×10^4	1.0×10^6
SPOILT FRUIT	3.5×10^7	2.5×10^6

Table 2 reveals the presence of the following above fungi isolated from the fruit head, bottom, internal part of the fruit, side/skin of fruit and the stalk.

Table 2: Fungi isolated from samples

PARTS	UNRIPE	RIPE	S POILT
Head	*Aspergillus niger, Mucor* spp	*Aspergillus niger,Mucor* spp *Geotrichum* spp	*Pencillium notatum, Aspergillus fumigatus, Torulopsis* spp, *Candida* spp, *Cryptococcus neoformans*
Bottom	*Torulopsis* spp, *Candida* spp, *Rhodotorula rubra* and *Aspergillus niger Aspergillus fumigatus, Candida tropicalis,. Candida quilliermondii*	*Cephalosporium* spp, *Geotrichum* spp	*Aspergillus fumigatus, Aspergillus niger, Candida tropicalis*
Internal	*Aspergillus fumigatus*	No growth	*Aspergillus, fumigatus, Candida tropicalis*
Side	*Aspergillus fumigatus, Aspergillus niger*	*Geotrichum* spp	*Trichosporon cutaneum, Candida pseudotropicalis*
Stalk	*Aspergillus fumigatus, Aspergillus niger*	*Trichosporon cutaneum*	*Aspergillus fumigatus, Aspergillus niger.*

Table 3 reveals the presence of the following bacteria; *Staphylococcus aureus*, and *Bacillus subtilis*.

Table 3: Bacteria Isolated From Samples

PARTS	UNRIPE	RIPE	SPOILT
Head	*Staphylococcus aureus*	*Bacillus subtilis*	*Staphylococcus aureus*
Bottom	*Staphylococcus aureus*	*Bacillus subtilis*	*Bacillus subtilis*
Internal	*Staphylococcus aureus*	*Staphylococcus aureus*	*Bacillus subtilis*
Side	*Staphylococcus aureus*	*Staphylococcus aureus*	*Bacillus subtilis*

Table 4 reveals that microbial contamination leads to fruits deterioration and consequently a reduction in the nutritional composition of *Chrysophyllum albidum* fruits especially in the spoilt sample.

Table 4: Nutritional Content Of *Chrysophyllum Albidum*

SAMPLE	MOISTURE	PROTEIN	CARBOHYDRATE	ASH	LIPID
Ripe	42.20%	8.01%	52.75%	26.2%	13.04%
Spoilt	30.2%	1.25%	74.07%	19.6%	5.08%
Unripe	38%	2.25%	69.13%	22.2%	6.42%

Discussion

Aspergillus fumigatus appeared to be the most prevalent of all the fungal isolates, followed by *Aspergillus niger* and *Geotrichum spp*. This observation agrees with the report of Oladiran *et al* (2002) who reported the prevalence of these organisms in soyabeans seeds in storage.

Oyeleke *et al.* (2002) reported that *Fusarium* spp, *Aspergillus fumigatus*, and *Trichophyton rubrum* are known to be associated with seed rot and sometimes produce aflatoxins. Thus, the spoilt fruits analysed in this study may have got rotten due to infestation by these fungi. The presence of these fungi species in the fruits may be due to infection from the field before storage. This agrees with Oyeleke *et al* (2002), which reported that, the longer the fruit stay un-harvested, the more the fungi load and this invariably affects the nutritional composition. This also agrees with Oladiran *et al* (2002) who reported that decline in seed viability may set in if harvesting is delayed after agronomic maturity. However, *Staphylococcus aureus* and *Bacillus subtilis* are most prevalent in both ripe, unriped and spoilt fruits. The prevalence of these organisms may be due to the favourable nutritional composition (Table 4) in accordance with Adelaja (1997) who reported an excellent source of carbohydrate, protein, fat and moisture in *C. albidum*.

This study showed that 65% of the fruits were infected. These infected fruits when packed with non-infected caused increased deterioration of Africa star apple (*Chrysophyllum albidum*) when in transit and storage (Adebisi, 1997). The natural dropping of star apple fruits from the height of the tree probably bruises the fruits and leads to entry of these fungi. It is also possible that insect vectors are involved in dissemination as reported by Adelaja (1997), indicating that fruit fly stings enhance the entry of *Geotrichum* spp into African star apple fruits by their oviposition on the fruits.

The high moisture content of the *Chrysophyllum albidum* fruit could also encourage colonization by moulds. Other factors that are responsible for fungi infestation of Africa star apple (*Chrysophyllum albidum*) includes temperature at which the fruits are stored. Shetty (1992) reported that *Aspergillus spp* and few *Penicillium* spp have ability to grow on grains seeds and fruits whose moisture level is in equilibrium with the relative humidity of 70-90%. Barney *et al* (1995) reported that surface disinfection was most effective in reducing abundance of *Penicillium* spp at low temperature and low moisture content. This may be the reason for the high infestation of the Africa star apple by fungi.

Following the percentage of nutritional contents obtained from the sample, it can be said that the fungi invasion has reduced the nutritional content of the spoilt fruits except for the carbohydrate content that increases as result of metabolic activities (Table 4). This is due to the microbial infestation of the fruit this agrees with the report of Adebisi, (1997) who reported a decrease in the nutritional content as a result of microbial contamination.

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