

Bio-Deterioration Of Shea Butter Fruit (*Vitellaria Paradoxa*) In Storage And Its Effects On The Nutrient Composition

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Abstract: The bio-deterioration of shea butter (*Vitellaria paradoxa*) in storage and its effect on the nutrients composition of the fruit was investigated in Ogbomosho, Southwestern Nigeria. Freshly dropped fruits were stored under laboratory for a period of 40 days. Eight fungi were isolated, which were *Aspergillus flavus*, *Aspergillus niger*, *Botrydiplodia theobromae*, *Botryosphaeria* spp., *Colletotrichum gleosporioides*, *Lisidiplodia* spp., *Pseudofasicocum* spp. and *Trichoderma viridae*. However, *B. theobromae* and *Lisidiplodia theobromae* were respectively the most predominant of all the isolates followed by *P. heterospora*, *Pseudofasicocum* spp. and *A. niger* while *T. viridae* was the least encountered. It is clearly revealed from the study that *A. niger*, *B. theobromae*, *Botryosphaeria* spp. and *P. theobromae* were highly pathogenic while *A. flavus*, *L. theobromae* and *Pseudofasicocum* spp. were moderately pathogenic. In addition, only *C. gleosporioides* were slightly pathogenic while *T. viridae* was non – pathogenic. It is clearly revealed from the study, that all the fungal isolates were isolated from the shea butter fruit throughout the 40DAS with the exception of *T. viridae*. Also, right from 30DAS, shea butter fruit inoculated with *A. niger*, *B. theobromae*, *Botryosphaeria* spp. and *P. theobromae* respectively developed extensive rotting with offensive odour. In addition, the result showed that the carbohydrate, moisture content, crude fibre, ash, ascorbic acid and energy respectively decreases significantly ($P \geq 0.05$) with increase in the days after storage while crude protein decrease significantly. Furthermore, all mineral contents analyzed decreased significantly ($P \geq 0.05$) with increase in days after storage. The significance of bio–deterioration of shea butter fruit in storage and its effects on the nutrient composition is here in discussed.

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Introduction

The shea tree recently has gained importance as an economic crop because of the soaring demand for its butter, both domestically and internationally. The shea tree also known as *Vitellaria paradoxa*, widely known under its synonym *Butryospermum paradoxum* belongs to the Sapotaceae family. It is considered a monotypic genus with two subspecies stretching nearly 5,000 km across the African savanna (Marnaz *et al.*, 2004; Fobil, 2010).

Shea fruits serve as an important source of food for many organisms and other animals including birds, bats, elephants, sheep and pigs (Marnaz *et al.*, 2004; USAID 2004; Fobil, 2010). The fruits also contribute to food security in forest areas of Nigeria, mainly for the rural poor, especially since their ripening happen together with the lean season of food production (Fobil, 2010). It plays a crucial role in its highly nutritional capacity, food security and availability during the period of low food availability. The production of shea nut and processing of the butter serves as one of the main sources of employment for the rural women and children who are engaged in gathering it. Shea butter is important edible oil for the

people of Southwestern Nigeria and most of Western Africa, being the most essential source of fatty acids and glycerol in their diet (Saul *et al.*, 2003; Moore, 2008). It is also useful in the pharmaceutical and cosmetic industries as an important raw material and a precursor for the manufacture of soaps, candles, and cosmetics (Fobil, 2010).

Shea nuts are also more and more being exported for the utilization in the cosmetics industry as a constituent in lotions, makeup, baby ointments, hair care products and soaps (Akosah-Sarpong, 2003; Moore, 2008). Regardless of being increasingly substituted by commercially produced lotions in many communities, shea butter is traditionally used as a skin and hair moisturizer and for protection against the sun (Ezema and Ogujiofor, 1992). The healing properties and effects of shea butter are thought to be somewhat attributable to the existence of allantoin, a substance known to trigger the growth of healthy tissue in ulcerous wounds (Wallace-Bruce, 1995). It is also traditionally smeared on pregnant women during childbirth, on new born babies and adolescents because of its relieving effects (Moore, 2008). Though the nuts being an essential export commodities, its

fruit pulp nevertheless is widely consumed. It also plays a major role in the local economy and diet as well as occupies an important period of time in the annual local dietary cycle (Maranz *et al.*, 2004). The edible part of the shea fruit is extremely nutritious and has important nutrients for the human body. The fruit provides important source of food for communities and rural poor particularly at the period of food shortages, hunger and other catastrophes. In addition to providing health benefits, it also provides some income (Okafor, 1985; FAO, 2007).

Fobil (2010) describes that the green fruit has a fleshy edible pulp, which contains 0.7-1.3g protein and 41.2g carbohydrate and is sweet. The pulp is also a rich source of ascorbic acid and has 196.1mg/100g compared with 50mg/100g in oranges. The iron and calcium content compares very well with raspberries: 1.93mg/100g as against 0.92mg/100g for iron, and 36.4mg/100g as against 26mg/100g for calcium (Fobil, 2010). Furthermore, FAO estimates that B vitamins are also present and the sugar content varies from 3-6%, equally distributed between glucose, fructose and sucrose (FAO, 1988). Okullo *et al.*, (2010) also analyzed the mineral composition and reported that, the fruit pulp contains significant and adequate amount crude proteins, crude fibre, vitamin C and nutrients (Ca, K, Mg, Na and Fe) and is equivalent to other edible fruits like mango and stated that it must therefore be promoted in human nutrition. Calcium intake lowers the risk of osteoporosis and gives strong bones (Van Camp *et al.*, 2009). Again deficiency in Fe can result in anemia especially in children and pregnant women. These potentials are good news in improving the health and nutritional status of individuals and communities with these health problems. Moreover, a similar study on the nutrient composition of the shea fruit pulp by Maranz *et al.*, (2004) showed many shea tree fruit pulp had potassium more than 1000mg/100g and has a rich source of calcium (141mg/100g) compared with marula (35mg/100g) and mango (10mg/100g). Despite the importance of shea butter fruit, its production is faced with several problems. This includes short shelf life and poor yield due to diseases (Cook, 1975; Fobil, 2010). Since the shelf lives of shea butter fruits are short, they are often utilized in Nigeria within 12 days of harvesting. The following fungi have been isolated from shea butter pulp rot (*Lasiodiplodia sp.*, *Botryosphaeria sp.*, *Pseudofusicocum sp.*, *Colletotrichum gloeosporioides*, *Pestalotiopsis sp.*, *Phoma sp.*, and *Phomopsis*) from most of the shea butter growing areas around the globe (Kieth *et al.* 2006; Sakalidis *et al.*, 2011). Most of these pathogens have also been reported from other parts of the world to cause shea butter fruit rot which could result in its fruit losses. However, it can take up to forty days

after harvesting to get to the markets in some major cities, resulting in huge losses due to bio-deterioration. This study was therefore designed to investigate the etiology of bio-deterioration of shea butter fruit in storage and the probable effect on its nutrient status.

Materials and Methods

Shea butter fruits were picked immediately after dropping from trees in a plantation at Ogbomosho, Oyo State in Southwestern Nigeria. Twenty-five trees were selected and three fruits per tree were used for this experiment. The fruits were kept in sterile polythene bags and taken to the microbiology laboratory of Pure and Applied Biology department, Ladoke Akintola University of Technology, Ogbomosho, Nigeria. After nine days of storage in clean chamber at room temperature, the fungi that emerged from the fruits were isolated aseptically and plated on sterilized Potato Dextrose Agar in Petri dishes.

The inoculated plates were then incubated at $25\pm 1^{\circ}\text{C}$ for 6 days. The associated pathogens were identified using cultural and morphological features and by comparison with culture which was obtained from the seed health pathology laboratory of the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria, and which had been identified by the International Mycological Institute, CABI Bioscience, Egham, UK.

Pathogenicity Test

Freshly harvested (matured) shea butter fruit were surface sterilized by swabbing with 70% alcohol and bored into with a sterile cork-borer (4mm in diameter). A similar sterile cork-borer was used to cut pellets of agar containing 3-day old cultures of fungal mycelia of the isolates. These fungi were then used to inoculate the hole created by scooping out shea butter fruit tissue. The scooped out tissues were replaced to cover the bored hole in the shea butter fruit. Three shea butter fruits were inoculated with each isolate and 3 inoculating site of equal distance on each fruit were used. The inoculated fruits were then enclosed in polythene bags containing moist cotton wool to maintain high relative humidity and incubated at 25°C in Gallenkamp incubators for seven days. Differences in the rate of infection were then recorded.

Determination of Moisture Content

Representative portion of the fresh shea butter fruit samples taken at intervals of two-day for 9 days were weighed (in triplicates). These same samples were placed in an oven at 80°C for 20 h, and weighed. The moisture content of the fruits was calculated according to AOAC (1984).

Nutrient Content Analysis

Three fruits from each tree were used for the analysis. Samples were excised from the fruits at a distance of 4cm from each other and at intervals of two days for nine days. These were weighed, cut into pieces and dried in a hot air oven at 60°C for 3 days. After grinding into powder, the samples (in triplicates) were analyzed for ash, crude fibre, crude protein, carbohydrate, energy and the nitrogen free extract using the official method of analysis (AOAC, 1984). Chemical elements were also analyzed according to standard methods (AACC, 1983) at the analytical laboratory of the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria.

Data analysis

Data are subject to the analysis of variance and means separated using Duncan Multiple Range Test (DMRT) at 5% level of probability.

Result

The result on the frequency of fungi associated with bio-deterioration of shea butter and their pathogenic importance during storage is presented in table 1. The result showed that nine fungi were found associated with softening of the mesocarp (pulp) of shea butter fruit leading to production and emitting of offensive odour. These fungi were *Aspergillus flavus*, *Aspergillus niger*, *Botrydiplodia theobromae*, *Botryosphaeria spp*, *Collectrichum gleosporoides*, *Lisiodiplodia theobromae*, *Pestalotia heterospora*, *Rhizopus stolonifer* and *Trichoderma viridae* (Table 1). However, *B. theobromae*, *L. theobromae* were respectively the most predominant of all the isolates followed by *Pestalotia heterospora*, *Pseudofasicocum spp*. (Table 1). It is clearly revealed from the study that *A. niger*, *B. theobromae*, *Botryosphaeria spp* and

Pestalotia theobromae were highly pathogenic while *A. flavus*, *L. theobromae* and *Pseudofasicocum spp* were moderately pathogenic. In addition, only *C. gleosporoides* were slightly pathogenic while *T. viridae* was non- pathogenic (Table 1). *B. theobromae* which occurred least in stored African star apple fruit has been reported to be one of the most important fruit pathogen in Southwestern Nigeria (Adisa and Fajola, 1982; Amusa *et al.*, 2003).

The incidence of occurrence of fungal isolates on shea butter fruit during storage is presented in Table 2. The result showed that until 5 Days After Storage (DAS), there was no occurrence of fungal isolates with the stored shea butter with the exception of *A. niger*, *A. flavus*, *L. theobromae*, *Pseudofasicocum spp* and *T. viridae* (Table 2). While all the fungi isolates were isolated from the shea butter fruit through the 40DAS with the exception of *T. viridae* (Table 2). The result clearly showed that right from 30DAS shea butter fruit inoculated with *A. niger*, *B. theobromae*, *Botryosphaeria spp* and *P. theobromae* respectively developed extensive rotting.

The freshly harvested shea butter fruit was found to contain carbohydrate and moisture contents of about 70.2% and 19.10%, respectively. The carbohydrate content decreases with storage time at room temperature, while increase in moisture was observed (Table 2). The fat content was low and continuous decrease was also observed during storage. The crude fibre, crude protein, and ash content of the fruits were found to increase with storage, while the energy of 310 KCal found in the freshly harvest shea butter fruit gradually decreased with increase in storage time. Generally, the analyzed minerals, which include potassium, iron, phosphorus, calcium and magnesium, increased with storage time (Table 3).

Table 1: Frequency of fungi associated with deterioration of shea butter and their pathogenic importance.

Fungi isolates	% Frequency*	Pathogenicity
<i>Aspergillus niger</i>	80 ^b	+++
<i>Aspergillus flavus</i>	41 ^c	++
<i>Botrydiplodia theobromae</i>	100 ^a	+++
<i>Botryosphaeria spp</i>	100 ^a	+++
<i>Collectotrichum gloeosporioides</i>	10 ^d	+
<i>Lisidiplodia theobromae</i>	100 ^a	++
<i>Pestaliopsis spp</i>	98 ^a	+++
<i>Pseudo fasicocum spp</i>	95 ^a	++
<i>Trichoderma viridae</i>	10 ^d	-

+ Slightly pathogenic

++ Pathogenic

+++ Highly pathogenic

*Data were arc sinned transformed before statistical analysis

Data in the same column with similar alphabet are not statistically different at 5% level of probability using Duncan Multiple Range Test (DMRT)

Table 2: The incidence of fungal isolates on shea butter fruit during storage

Fungal isolates	Incidence of occurrence at different days after storage								
	0	5	10	15	20	25	30	35	40
<i>Aspergillus niger</i>	-	-	+	+	+	+	+	+	+
<i>Aspergillus flavus</i>	-	-	+	+	+	+	+	+	+
<i>Botryodiplodia theobromae</i>	-	-	+	+	+	+	+	+	+
<i>Botryosphaeria spp</i>	-	-	+	+	+	+	+	+	+
<i>Colletotrichum gloeosporoides</i>	-	-		+	+	+	+	+	+
<i>Lisideplodia theobromae</i>	-	-	+	+	+	+	+	+	+
<i>Pestaliopsis spp</i>	-	-		+	+	+	+	+	+
<i>Pseudo fasciocum spp</i>	-	-	+	+	+	+	+	+	+
<i>Trichoderma viridae</i>	-	-	+	+	+	+	+	+	+

+ = Present

- = Absent

Table 2: Effect of Bio-deterioration of shea butter fruit in storage in its nutrient composition

Days in storage	Nutrient composition							
	Carbohydrate (g/100g)	Moisture content (g/100g)	Fat (g/100g)	Crude fibre (g/100g)	Crude protein (g/100g)	Ash (g/100g)	Ascorbic acid (g/100g)	Energy (g/100g)
1	43.00 ^a	03.80 ^a	34.53 ^a	12.00 ^a	01.60 ^a	05.20 ^a	196.10 ^a	248.16 ^a
5	42.06 ^a	03.61 ^a	33.01 ^a	11.54 ^a	01.60 ^a	05.20 ^a	196.10 ^a	248.16 ^a
10	38.51 ^{ab}	03.26 ^a	30.70 ^b	11.00 ^a	01.61 ^a	05.20 ^a	196.10 ^a	248.16 ^a
15	32.70 ^c	03.00 ^a	29.01 ^b	08.25 ^b	1.40 ^{ab}	04.90 ^b	190.00 ^a	240.00 ^b
20	25.30 ^d	02.80 ^b	25.17 ^c	06.00 ^c	1.25 ^b	04.71 ^c	171.00 ^b	235.01 ^c
25	20.61 ^e	02.25 ^c	21.00 ^d	04.25 ^d	1.10 ^b	04.53 ^d	160.00 ^c	230.13 ^d
30	15.08 ^f	01.80 ^d	17.53 ^e	03.00 ^d	0.80 ^c	03.51 ^e	150.00 ^d	223.00 ^e
35	10.00 ^g	01.51 ^e	12.00 ^f	02.00 ^{de}	0.61 ^d	03.00 ^f	135.00 ^e	215.11 ^f
40	05.71 ^h	01.00 ^f	07.17 ^g	01.00 ^e	0.40 ^e	02.63 ^g	120.00 ^f	209.00 ^g

Data in the same column with similar alphabet are not statistically different at 5% level of probability using Duncan Multiple Range Test (DMRT)

Table 3: Effect of Bio-deterioration of shea butter fruit on its mineral contents.

Days in storage	Mineral composition				
	Iron (g/100g)	Calcium (g/100g)	Magnesium (g/100g)	Potassium (g/100g)	Sodium (g/100g)
1	03.00 ^a	36.00 ^a	27.00 ^a	42.31 ^a	07.07 ^a
5	03.00 ^a	36.00 ^a	27.00 ^a	42.31 ^a	07.07 ^a
10	03.00 ^a	36.00 ^a	27.00 ^a	42.31 ^a	07.07 ^a
15	02.71 ^b	34.50 ^b	25.01 ^b	40.35 ^b	06.11 ^{ab}
20	02.20 ^b	30.00 ^c	22.71 ^c	36.00 ^c	05.00 ^b
25	02.00 ^b	25.78 ^d	20.00 ^d	30.13 ^d	03.06 ^{cd}
30	01.60 ^c	20.11 ^c	17.11 ^c	29.81 ^c	02.10 ^{dc}
35	01.20 ^d	15.00 ^f	12.00 ^f	20.00 ^f	01.15 ^c
40	01.00 ^d	10.00 ^g	08.65 ^g	15.06 ^g	0.41 ^f

Data in the same column with similar alphabet are not statistically different at 5% level of probability using Duncan Multiple Range Test (DMRT)

Discussion

The fungi found associated with deteriorating shea butter fruit in storage were *Aspergillus flavus*, *Aspergillus niger*, *B. theobromae*, *Botryosphaeria spp*, *Collectotrichum gleosporoides*, *L. theobromae*, *Petaliopsis spp*, *Pseudofusicocum spp* and *T. viridae*. Purseglove (1968) reported that *Rhizopus artocapi*, which is similar to one of the fungus observed in this study, as responsible for the soft rots of shea butter fruit in India. While the fungi found associated with the deteriorating seeds of African shea butter fruit (*Treculia africana*) in Nigeria are *A. niger*, *R. stolonifer*, *B. theobromae* and yeast (Nwufo, and Mba, 1987).

The decrease in carbohydrate content of shea butter fruit stored at room temperature might be due to fermentation caused by microbes and the respiratory loss of sugars as CO₂. Parkison (1984) have also reported that fermented shea butter fruit is low in carbohydrate content. Ikediobi and Oti (1983) as well as Ravinduram and Wanasindera (1992), also attributed the steady decline in starch contents of stored *Dioscorea rotundata* tubers to the respiratory loss of sugars as carbon dioxide. There was also fat decrease in shea butter fruit with storage time, which is consistent with the observation that fermented shea butter fruit is low in fat as compared to fresh shea butter fruit (Nwufo and Mba, 1987; Thompson et al., 1974).

The increase in moisture content with storage time might partly be due to metabolic water. Ladele *et al.* (1984) had reported that the moisture content of banana increased during storage, while the moisture content of ripe banana is higher than that of the unripe fruit (Ketiku, 1973). Increase in crude fibre content was observed as from the 5th day. Awan and Ndubizu (1978) earlier observed that fibre content remained almost constant for the first week of storage. Ketiku (1973) however reported an increase in fibre contents from 0.5% - 1.1% in the unripe banana to ripe pulp. Our observation of the increase in ash during storage is in agreement with the report of Ketiku (1973) who showed that ash content of banana increased from 2.0g in unripe plantain to 2.2g in the ripe fruit. The significant decrease in carbohydrate content inocula shea butter samples could result from the utilization of carbohydrate by the mould metabolism (Onions *et al.*, 1981, Oyeyipo *et al.*, 2011). Also the increase in protein contents of shea butter fruit with increase in storage time is similar to the observation of other fruits (Ketiku, 1973; Awan and Ndubizu, 1978) the later reported an 89.9% increase in total protein content of African pear following inoculation with *Aspergillus niger*. These observations may be partially attributed to the utilization of products of carbohydrate degradation for protein synthesis by the

fungus. It is therefore advocated that shea butter fruit pulp be utilized within the first-four days after harvest. This will not only prevent excessive infection of the pulp by fungal pathogens but will also eliminate the possibilities of contamination with mycotoxins and other related metabolites of infecting pathogens that might be hazardous to human health.

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