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# Mycological, Proximate and Mineral Analysis of Coated and Decoated Seeds of Fluted Pumpkin (*Telfairia occidentalis [F]* Hook)

**Dr. Emiri U. N.**
Senior Lecturer, Department of Agricultural Education,
Isaac Jasper Boro College of Education, Bayelsa State, Nigeria
**Enaregha E.**
Senior Lecturer, Department of Biology,
Isaac Jasper Boro College of Education, Sagbama, Bayelsa State, Nigeria

***Abstract:***
*Comparative studies on mycological, proximate and mineral analysis of coated and decoated seeds of fluted pumpkin (Telfairia occidentalis(F) Hook) were carried out. Results showed that four genera of fungi (Rhizopus, Aspergillus, Fusarium and Penicillium) were isolated from coated and decoated seeds. Rhizopus and Aspergillus were predominant in both seed samples. Decoated seeds harboured more fungi species; R. stolonifer 40%, A. niger 50%, A. flavus 60% and F. oxysportum 20%. While R. stolonifer 80%, A. niger 26% and P. digitatum 18% were isolated from coated seed samples. Proximate composition for coated and decoated seeds revealed moisture (20.1 and 22.5%), Ash (3.25 and 3.45%), Fibre (7.52 and 3.65%), Lipid (25.1 and 28.95%), Carbohydrate (12.82 and 10.87%) and Protein (31.21 and 30.58%) respectively. Mineral composition comprising of magnesium, potassium and sodium were higher in decoated seeds, while calcium, phosphorus and iron were higher in coated seeds.*

***Keywords:*** *Telfairia occidentalis, coated, decoated, seeds, fungi, proximate composition*

## 1. Introduction

Vegetables are associated with both normal flora, human and animal enteric pathogens except the soil-borne spore formers such as *Clostridium perfringes* and *Bacillus cereus* which are usually absent from fresh vegetables (Montville and Matthew, 2008). Notable among these vegetables is *Telfairia occidentalis.*

*Telfairia occidentalis* otherwise called fluted pumpkin (Ugu in Igbo) belongs to the cucurbitacea family (Ehiagbonare, 2008). The fluted pumpkin is mostly cultivated by women in the Eastern part of Nigeria. It can be cultivated on the flat land or on mounds. *Telfairia* does best at the lower attitudes and medium to high rainfall locations and will do well on sandier soil provided fertilizers are applied but have a more robust growth in rich and well-drained soil. It is known to have various nutrients such as fiber, carbohydrates, protein, ash, fat and other essential minerals. Consumption of this vegetable is therefore very beneficial (Eberhardt,*et al.,* 2000; Yusuf *et al.,* 2006; Idris, 2011). The crop not only provides nutrients to the body but also serves as medicine in a variety of ailments such as diabetes, colon cancer, lowers plasma cholesterol and softens stools (Gordon, 2000; Luc 2004; Ekeanyanwu *et al.,* 2010; Gafar *et al.,* 2010).

The young leaves are sliced and stored in a bottle to which coconut water and salt are added and used for the treatment of convulsion in ethno medicine (Ehigonare, 2008). The leaves contain essential oils and vitamins while root contains Cucubitacine, sequisterpene and lactones. The leaves are rich in iron and play a key role in the cure of anemia. The leaves are also noted for lactating properties and are in high demand for nursing mothers. The leafy vegetable is used mainly for soups and salads to accompany main course (Hopkins, 2001; Agatemor, 2006 a). The roots are used as rodenticide and ordeal poison.

The seed of fluted pumpkin (*Telfairia occidentalis*) is widely consumed in Nigeria, especially in the South Eastern part of Nigeria where it is used as a condiment in soup. The fermented seeds of fluted pumpkin are used in the production of "Ogiri ugu", a locally made custard. They could also be used in cookie formulations and marmalade manufacturing (Giami and Barber 2004).

The microbial and proximate analysis of the leaves and seeds has been reported (Lennox and Nkra 2016). However, the mycological, proximate and mineral analysis of coated and decoated seeds of *Telfairia occidentalis* have not been reported. This has informed the decision to carry out this research; to identify the fungi associated with the coated and decoated as well as investigate their proximate, mineral and phytochemical compositions.

## 2.Materials and Methods

*2.1. Collection of Samples*

Coatedand decoated *Telfairia occidentalis (*Pumpkin) seeds were purchased from oil mill market in Port Harcourt, Rivers State, Nigeria.

The samples were taken to the Plant Pathology laboratory for further studies.

*2.2. Proximate Composition Determination*

The samples of Pumpkin seeds were taken to the laboratory for the determination of their proximate compositions comprising of ash, moisture, fibre, lipid, carbohydrate and protein, as well as their mineral content. These parameters were determined according to the method of Association of Official Analytical Chemists (AOAC, 1990).

*2.3. Media Preparation*

The medium used for fungal isolation was the Sabouraud Dextrose Agar (SDA). This was prepared by weighing 32.8g of Sabouraoud Dextrose Agar (SDA) into a 500ml conical flask. Distilled water (500ml) was added into the flask with a measuring cylinder and stirred to homogenize. The mouth of the conical flask was plugged with sterile cotton wool and wrapped with foil. The conical flask with its contents was autoclaved for 15 minutes at 121$^{o}$C at 1.1kg cm$^{-3}$ pressure. Sterile petri dishes were prepared and the mixture dispensed into them while still hot and allowed to solidify.

*2.4. Mycological Studies*

2.4.1. Isolation and Identification of Fungi

Three seeds of *T. occidentalis* used were washed in tap water, rinsed in distilled water and surface sterilized with 5% Sodium hypochlorite and rinsed twice in sterilized distilled water after which they were aseptically introduced into the SDA in petri dishes equidistantly, in triplicate. The inoculated plates and their contents were incubated for 7 days at room temperature of 28 ± 2$^{o}$C. Pure culture of fungi growing in mixtures was obtained thereafter.

Pure cultures of the isolates were made after series of isolation. The fungi were later identified using fungi identification guides by Barnett and Hunter (1999) and Alexopoulus *et al.,* (2002)

2.4.2. Pathogenicity Studies

Healthy samples of coated and decoated *T. occidentalis* seeds were washed in tap water and surface sterilized in 5% sodium hypochlorite. The fungi isolates were aseptically inoculated onto the healthy seeds on damp blotter papers in petri dishes and incubated at room temperature of 28 ± 2$^{o}$C for five days. Petri dishes containing seeds of *T.occidentalis* without the fungal isolates served as control. The extent of rot was determined using the method as described by Agrios (2005) and Trigiano *et al.,* 2004.

2.4.3. Mean percentage Incidence of Fungi

The mean percentage incidence of fungi was calculated using the formula:

$$\text{Mean Percentage} = \frac{\textit{Total number of occurence of a particular fungi}}{\textit{Total number of plated sample}} \times \frac{100}{1}$$

## 3.Results

Results on the incidence of fungi isolated from coated and decoated *T. occidentalis*seeds are presented in table 1.

| Fungal Isolates | Coated | Decoated |
|---|---|---|
| *Rhizopus stolonifer* | 80 ± 0.15 | 40± 0.20 |
| *Aspergillusniger* | 26 ± 0.22 | 50 ± 0.28 |
| *Aspergillusflavus* | - | 60 ± 0.33 |
| *Fusariumoxysporum* | - | 20 ± 0.30 |
| *Penicilliumdigitatum* | 18 ± 0.25 | - |

*Table 1: Mean percentage incidence of fungi isolated from coated and decoated seeds of T. occidentalis*

Results reveal that *Rhizopus stolonifer* and *Aspergillusniger* were predominant in coated and decoated *T. occidentalis* seeds. Decoated seeds harboured more fungal species. *Aspergillusniger* and *Fusariumoxysporum* were found only in decoated seeds while *Penicilliumdigitatum* was found only in coated seeds.

Results on proximate and mineral compositions are presented in table 2 and 3 respectively.

| Parameter | Coated Seeds (Values %) | Decoated Seeds (Values %) |
|---|---|---|
| Moisture | 20.1 ± 0.20 | 22.5 ± 0.25 |
| Ash | 3.25 ± 0.10 | 3.45 ± 0.22 |
| Fibre | 7.52 ± 0.10 | 3.65 ±0.15 |
| Lipid | 25.1 ± 0.22 | 28.95 ± 0.03 |
| Carbohydrate | 12.82 ± 0.32 | 10.87 ± 0.12 |
| Protein | 31.21 ± 0.20 | 30.58 ± 0.30 |

*Table 2: Proximate composition of coated and decoated seeds of T. occidentalis*

Results revealed that Protein, Lipid and moisture contents were higher than Carbohydrate, Fibre and Ash.
Comparing coated and decoated samples, the contents of Proteins, Carbohydrate and Fibre were higher in coated seed samples while moisture, Ash and Lipids were higher in decoated samples.

| Parameter | Coated seeds (Values %) | Decoated seeds (Values %) |
|---|---|---|
| Calcium | 0.6 ± 0.08 | 0.58 ± 0.04 |
| Phosphorus | 0.05 ± 0.04 | 0.04 ± 0.03 |
| Sodium | 0.01 ± 0.01 | 0.02 ± 0.01 |
| Potassium | 0.57 ± 0.14 | 0.58 ± 0.10 |
| Iron | 0.53 ± 0.10 | 0.52 ± 0.08 |
| Magnesium | 4.25 ± 0.20 | 4.44 ± 0.22 |

*Table 3: Mineral composition of coated and decoated seeds of T. occidentalis*

Results showed that magnesium was relatively high. The value of magnesium, Potassium and Sodium were higher in decoated samples while Calcium, Phosphorus and Iron were higher in coated sample. Though differences in value were not significant.

| Parameter | Coated Seed (Values %) | Decoated Seeds (Values %) |
|---|---|---|
| Tannin | 0.05 ± 0.02 | 0.05 ± 0.02 |
| Total Oxalate | 0.05 ± 0.02 | 0.02 ± 0.01 |
| Hydrogen cyanide | 0.00 ± 0.00 | 0.00 ± 0.00 |

*Table 4: Phytochemical of coated and decoated C. colocynthis seeds*

Results showed that *T. occidentalis* seeds lack hydrogen cyanide. Tannin had the same value in both samples, while total oxalate was higher in coated seeds.

## 4. Discussion

*4.1. Percentage Incidence of Fungi*

Fungi have been recorded as accounting for up to 95% and bacterial 5% loss of fluted pumpkin fruits during storage under ambient conditions (Odiaka and Schippars, 2004). Four genera of fungi (*Rhizopus, Aspergillus, Fusarium* and *Penicllium* were isolated from *T.occidentalis* seeds. This agrees with the assertion of Lennox and Nkra (2016). *Rhizopus*and *Aspergillus* species were predominant in both seed samples. It therefore suggests, that *Rhizopus* and *Aspergillus* species are more abundant in the air/environment (Ueno, 2000). Decoated seeds harboured more fungi than coated seeds. This was expected because seeds are generally protected by differential integumentary structures (seed coat) which serves as barriers to microbial invasion. Therefore, when the seed coat is removed, the barrier is broken, hence the microbial invasion. This could also be attributed to the method of handling and processing. It has been reported that the method of processing, handling and preservation affects the level of contamination and influence of microbial load of agricultural products. (Chukwu *et al.*, 2009; Etebu and Emiri, 2016). All the fungal isolates from coated and decoated *T. occidentalis* seeds were found to be pathogenic causing general soft rot of seeds leading to the decay of the seeds and loss of quality.

*4.2. Proximate Composition of Coated and Decoated T. occidentalis Seeds*

The moisture content ranged between 20.1 and 22.5 for coated and decoated seeds respectively. (Table 2) These values were high and will encourage deterioration due to microbial attack, hence, the high fungi incidence particularly in decoated samples. The moisture content was lower in coated seed samples; this was anticipated given the dry nature of the seed coat. The value of moisture was significantly higher than 10.93% reported by Agatemor (2006) on coated *T. occidentalis* seeds. This variation could be attributed to the method of processing and preservation.

The ash content was low; 3.25 and 3.45 for coated and decoated seeds respectively. It is comparable to the result of Lennox and Nkra (2016) on coated fluted pumpkin seeds. Measure of ash content could be a measure of food quality. The ash content from this work was however significantly higher the report of Agatemor (2006) on the same seed.

Fibre was also investigated. It ranged from 7.52 and 3.65% for coated and decoated seeds respectively. The values were comparable to 3.44 reported for coated seeds of *T. occidentalis* (Lennox and Nkra 2016). Fiber regulates bowl actions and my help to guard against colon and rectal cancer as well as in diabetes. Fibre shortens the transit time of food through the gastrointestinal tracts, reduces low density lipoprotein and hence keeps the gut healthy.Fibre supplements or fibre-rich foods may function as normal dietary agents by modulating the digestive and absorptive process (Okaka *et al.,* 2006).

The percentage compositions of lipids in the samples were relatively high. The lipid contents for coated and decoated seeds were 25.1 and 28.95% respectively. The values were lower than 33.38% and 39.80% reported by Agatemor (2006), Lennox and Nkra (2016) respectively for coated seeds. These variations in the lipid contents may be attributed to differences in climatic conditions, soil properties, average rainfall, freshness and storage conditions / time of the seeds. Lipids are the principal form of stored energy (fats and oils) in most organisms and major constituents of cellular membranes. The lipid content indicates that the seed is a potential source of edible oils.

Coated and decoated seeds of *T. occidentalis* had 12.82% and 10.87% carbohydrate composition respectively. These values were low and agree with the assertion of Lennox and Nkra (2016) on the same seed.

*T. occidentalis* seeds contained an appreciable amount of protein. The composition of protein ranged from 31.21 and 30.58% for coated and decoated seed samples respectively. The values were however; lower than the results of Lennox and Nkra (2016). The decrease could be as a result of the processing method in the preparation of the seed samples and other environmental factors. These values give the seeds positive attributes as plant proteins are scare and this protein content can furnish the essential amino acids needed for healthy growth and repair of tissue (Igwenyi, 2008).

*4.3. Mineral Composition of Coated and decoated T. occidentalis seeds*

Minerals are important in the diet because they serve as cofactors for many physiologic and metabolic functions. *T. occidentalis* seeds contained levels of important minerals, (Table 3) which when consumed in adequate amounts could satisfy about one third of the daily requirement of this minerals. Calcium, a vital mineral for bone and muscle growth, as well as neurological function, was present in the seed. The daily requirement of calcium is 1200mg until the age of 24 years. So an adequate consumption *Telfairia occidentalis* (approximately 1kg) per day would satisfy about one-fourth of this requirement (NRC, 1989).

Phosphorus is another important mineral for which deficiency could result in rickets (Scariona *et al.,* 1995).The seed contained some amounts of phosphorus, which could supply more than half of the daily requirement of this mineral, if the seed is severed in modest quantity.

Iron which is required for hemoglobin formation, was detected, though in little amount. Magnesium, another vital mineral was present at levels at which 500g serving per day would supply more than the body daily requirement.

Sodium, a principal cation in the extra-cellular medium was also checked, though in very low amounts. Potassium was also detected, though very low. The difference in mineral composition between coated and decoated seed samples was insignificant.

*4.4. Phytochemicals (Anti-nutrients) of Coated and decoated T. occidentalis Seeds*

Phytochemicals are secondary metabolic compounds from plants (Adodo, 2002). Results on phytochemicals (Table 4) showed that *T. occidentalis* seed did not contain hydrogen cyanide. Tannin and Oxalate were present, though very low. Tannins are known to possess health benefits, wherein they are 15 – 30 times more efficient in free radical quenching activity than Trolox and other simple phenolics (Hurrel *et al.,* 1999). They are astringent, bitter plant polyphenols that either bind and precipitate or shrink proteins. Recent studies have demonstrated that products containing chestnut tannins include at low dosages (0.15 – 0.2%) in the diet can improve wellbeing (Schiavone *et al.,* 2007).

## 5.Conclusion

Four genera of fungi (*Rhizopus, Aspergillus, Fusarium* and *Penicillium*) were isolated from coated and decoated seeds of *T. occidentalis*. Decoated seeds harboured more fungi species, *T. occidentalis* seeds contained high moisture content which made it vulnerable to fungal attack. The lipid content indicates that the seed is a potential source of edible oil. The protein content is also an indication that they can provide amino acids needed to support the metabolic activities of the body. The seeds also contained some mineral content and phytochemicals.

However, the effect of *mycodeterioration* on the proximate composition of *T. occidentalis* is advocated.

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