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Nutrient Content and Associated Mycoflora of Cola Parchycarpa (Monkey Kola) Sold in Port Harcourt Metropolis, Rivers State Nigeria.

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Abstract: *Studies on fungal pathogens and nutrient composition of Cola parchycarpa (monkey kola)was carried out in the laboratories of the Departments of Plant Science and Biotechnology and Food Science and Technology in Rivers State University. Proximate parameters assessed were moisture, ash, fibre, lipid, carbohydrate and protein. Moisture and protein had higher values 60.5±0.015% and 17.56±0.015% repectively for the spoilt fruit samples while ash, fibre and carbohydrate were higher for the healthy fruit samples (2.5±0.012%, 1.25±0.014% and24.88±0.033). Lipidcontent was the same (0.5±0.021) for both wholesome and spoilt fruit samples. Calcium, iron, magnesium, potassium, phosphorus, sodium and Vitamin C were the minerals and vitamin present in the fruits of C. pachycarpa. Calcium content was10.50±0.012mg/100g for both healthy and spoilt samples of C. parchycarpa. Higher values were also recorded for magnesium, potassium, phosphorus and sodium for the spoilt samples. Iron and vitamin C contents were higher for the healthy fruit samples. Phytochemical investigation revealed the presence of tannins, saponins, oxalate and cyanogenic glycoside in trace amounts. Four fungi were isolated viz: Fusarium moniliforme, F. oxysporum,Aspergillus niger and A. flavus and they all proved to be pathogenic to healthy fruits of C. Parchycarpa causing general soft rot of the fruits. However, A. flavus had the highest percentage incidence (40%). This was followed by F. moniliforme (30%) and F. oxysporum(20%) while A. niger had the least incidence (10%).*

Key word: *Cola parchycarpa, fungal pathogen, nutritional composition and monkey kola*

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I. Introduction

Cola parchycarpa commonly known as the yellow monkey kola is a member of the taxon Malvaceae (family) and Sterculioideae (sub-family) (APG, 2009). The plant is tropical and indegenous to Africa. It is widely distributed in West Africa in countries like Nigeria and Cameroon (Burkill, 1985). It has also been reported that Southern Nigeria could be referred to as the primary domestication center of species belonging to the monkey kola as they posses abundant species variety of the Cola genus (Anya, 1982; Ogbu and Umeokechukwu, 2014). Monkey kola also has other variety namely C. lepidota (white kola) and C. latertia (red kola) (Meregini, 2005; Ogbu et al., 2007).C. parchycarpa fruits can attain a lenght of 8.70 to 25.90cm and a thickness of about 3.65 to 5.53cm (Ogbu and Umeokechukwu, 2014).

Early studies on C. parchycarpa and other Cola spp have shown them to have nutritional, medical and industrial importance (Singh et al., 2010; Iguodala & Lanre, 2018; Ene-Obong et al., 2010; Giwa et al., 2012). Essien and Imabong, (2017) reported thatC. parchycarpa fruits contain moisture, protein, fibre, ash, lipid and carbohydrate in varying quantities. This was supported by the research of Okudu et al., (2015) as they also reported these macro nutrients in C. parchycarpa juice. Monkey kola has also been documented by early findings to contain several minerals and vitamins especially calcium, magnesium, sodium, potassium, phosphorus, iron, Vitamin C, β carotene, niacin, thiamin and riboflavin in the fruit and juice(Okudu et al., 2015; Essien and Imabong, 2017). The occurrence of these nutrients, minerals and vitamins have also been reported forC. lepidota (Imabong & Essien, 2017).

Several phytochemicals have been reported to be present in pulp and juice of monkey cola. These include, cyanide, tannins, phytates, oxalates, saponins and alkaloids (Okudu et al., 2015; Essien & Imabong, 2017). This was also supported by the investigation of Essien et al., (2015) as they were able to show the above phytochemicals in C. rostrata and C. lepidota. These phytochemicals in the seed and pulp extract of C. millenii have been shown to have antimicrobial potentials (Giwa et al., 2012). In addition, the research of Okudu & Asumugha, (2018) also confirms the presence of these phytochemicals as well as the above mentioned nutients,

minerals and vitamins in the seeds of C. parchycarpa obtained from South-east and South-south regions of Nigeria.

However, little is known about the mycoflora and possible spoilage fungi of monkey kola fruit, although the research of Okudu and Ene-Obong, (2015) showed that C. parchycarpa jam and juice had plate count of microorganisms (42.5cfu/ml) after four weeks storage. This report is an indication that there are the possibilities that microorganisms are associated with the spolage of these fruits used in juice and jam production. This experiment was designed to evaluate the nutrient quality and associated mycoflora of C. parchycarpa sold within Port Harcourt metropolis of Rivers State,Nigeria. The knowledge of which would proffer suggestions for proper handling and preservation methods of this fruit as well as highlight the nutritional benefits of its consumption .

II. Materials and Methods

Sample Collection

Samples of healthy and rotted fruits of C. parchycarpa and partially rotted fruits were bought from the local traders from Mile 3 market in Diobu Port Harcourt and brought to the Department of Plant Science and Biotechnology and sent to the Plant Pathology Laboratory for futher studies.

Determination of Nutrient composition

Healthy fruit samples were taken to the laboratories of the Department of Food Science and Technology for proximate, mineral and other phytochemical analysis using the AOAC (2012)standard methods of analyses.

Mycological studies

Preparation of mycological medium

Sterilization of conical flask, slides, Petri dishes and all the equipment needed for the research were carried out in the laboratory. The glassware were sterilized in the oven at 160^{o}C for 2h after washing with soap, while other equipment were surface sterilized with 70% ethanol. (Agrios, 2005). Inoculating loops and scalpels were sterilized by dipping for 20 seconds in 70% ethanol and heated to red hot. Sabouraud Dextrose Agar was prepared using the standard method. The mouth of the flask was plugged with non-absorbent cotton wool and wrapped with aluminium foil. The conical flask containing the mycological medium was autoclaved at 121^{o} C and pressure of 1.1kg cm-3 for 15 minutes. The molten agar was allowed to cool to about 40 ^{o} C and dispensed into Petri dishes at 15mls per plate and allowed to further cool and solidify.

Isolation of fungi from partially rotted C. parchycarpa fruits.

One gram of sample showing visible signs of spoilage by moulds was cut from the healthy portions of the fruits up to the points where rot had been established and inoculated onto Sabouraud dextrose agar with ampicillin(to hinder the growth of bacteria) in Petri dishes .Plating was done in triplicate and the inoculated plates were incubated for 5 days at ambient temperature of 25^{o} C ± 3^{o} C (Baudoni, 1988, Chuku, 2009, Samson et al, 1981). The entire set up was observed for 7 days to ensure full grown organisms. Pure cultures of isolates were obtained after a series of subcultures on fresh sterile medium.

Identification of fungi associated withC. parchycarpa.

Microscopic examination of fungal isolates was carried out by the needle mount method (Cheesebrough, 2000). The fungal spores were properly teased to ensure proper separation and visibility. The evenly spread spores were stained with cotton blue in lactophenol and examined microscopically using low and high power objectives. The fungi were identified based on their spore and colonial morphology, mycelia structure and other associated structures using the keys of (Samson et al, 1981 and Olds, 1983).

Pathogenicity studies

Pathogenicity studies were carried out on C. parchycarpa to check if the fungi isolated from the rotted fruits were capable of causing spoilage on healthy fruit samples. The methods of (Agrios, 2005, and Trigiano, 2004) were basically followed. The fungal isolates were innoculated into healthy fruits and incubated for seven days. The set up was examined visually daily for growth and signs of spoilage**.**

III. Result And Discussion

Table 1: Proximate composition of healthy and spoilt C. parchycarpafruits.

Parameters	Healthy (%)	Spoilt (%)
Moisture	55.5± 0.016	60.5±0.015
Protein	15.37±0.014	17.56±0.015
Lipid	0.50 ±0.021	0.50±0.021
Ash	2.5 ± 0.012	2.0±0.023
Fibre	1.25 ±0.011	1.05±0.013
Carbohydrate	24.88±0.033	18.39±0.022

Table 2: Minerals and vitamin composition of healthy and spoilt C. parchycarpafruits.

Parameters	Healthy (mg/100g)	Spoilt (mg/100g)
Calcium	10.50± 0.012	10.50±0.021
Iron	0.40±0.022	0.39±0.023
Magnesium	8.81±0.013	8.82±0.016
Potassium	1.05±0.011	1.06±0.014
Phosphorus	8.81±0.018	8.85±0.025
Sodium	0.99±0.031	1.02±0.001
Vitamin C	55±0.022	50±0.013

Table 3: Phytochemical composition of C. parchycarpa

Parameter	Percentage occurrence (%)
Tannin	0.4±0.05
Saponin	0.1±0.02
Oxalates	0.21±0.01
Cyanogenic glycoside	2.0±0.04

Table 4: Fungi isolates and percentage incidence

Isolates	Percentage incidence (%)
Fusarium moniliforme	30%
Fusarium oxysporum	20%
Aspergillus niger	10%
Aspergillus flavus	40%

The result of proximate analysis presented in Table 1 shows that C. parchycarpa had lower value of moisture (55.5 ±0.016) for the healthy samples but higher (60.5±0.015) for the spoilt fruit samples. The lipid content was equal for both healthy and spoilt samples. Nevetheless, higher values (2.5± 0.012, 1.25±0.011 and 24.88±0.033) were recorded for ash, fibre and carbohydrate respectively in the healthy fruit samples. However, protein value was higher in the spoilt samples. The may be atributed to to increased microbial biomass with spoilage. Moisture, protein , fat,ash and crude fibre contents obtained in this study are higher than those reported by Okudu & Asumugha, (2018) in the seed of C. parchycarpa. However, the value for moisture recorded for the healthy fruit samples in this study agrees with that(55.00±0.50) reported by Imabong and Essien, (2017) for the fruit pulp of C. lepidota. These authors also reported higher values for ash, fibre and carbohydrate though their protein value was lower compared to its equivalent in this study. The report of Okudu et al., (2015) showed lower values for all proximate parameters examined in this study excluding moisture for C. parchycarpa.

Table 2. shows the mineral and vitamin composition of C. parchycarpa. Calcium content was the same (10.50 ± 0.012mg/100g) for both healthy and spoilt fruit samples. However, higher valuesof magnesium (8.82 ± 0.016mg/100g), potassium (1.06 ±0.014mg/100g), phosphorus (8.85 ±0.025) and sodium (1.02 ±0.001) were obtained for the bad fruit samples compared to the values obtained for the wholesome samples. Iron and Vitamin C values for healthy fruits, 0.40± 0.022ng/100g and 55 ± 0.022 respectively are higher than those obtained for spoilt C. parchycarpa fruits. Minerals and vitamins perform essential metabolic roles in man and other organisms however, values reported in this study indicate thatC. parchycarpa may not be a good source of them.Thus the reccomended daily intake for calcium is 1000mg per day. Calcium content in this study is 0.1%. The values of these parameters in this study disagrees with that reported by Okudu et al., (2015) in C. parchycarpa juice as they are higher compared those in of this study. Also, the values reported by Okudu and Asumugha, (2018) for the seeds were all higher than those recorded in this study. Notwithstanding, the vitamin C value in this study is higher than that earlierly reported by Imabong and Essien, (2017) and Essien and Imabong, (2017) for C. lepidota and C. parchycarpa respectively. It wasreported earlier that C. lepidota had higher values of mineral constituents compared to C. parchycarpa (Imabong & Essien, 2017).

The result of phytochemical investigation is shown in Table 3 and reveals the presence of tannin, saponin, oxalate and cyanogenic glycoside in trace amount. These substances are in line with those reported earlier by other researchers to be present in C. parchycarpa and its relatives (Iguodala et al., 2018). This is supported with the investigation made by Essien et al., (2015) as they reported the presence of all the phytochemicals stated above, and many others in C. rostrata and C. lepidota. The research of Okudu and Asumugha, (2018) and Essien and Imabong, (2017) further confirms the presence of these phytochemicals in C. parchycarpa. The potential of these phytochemicals to inhibit microorganisms have been investigated and found to be effective. The research of Adewumi and Arije, (2017) showed that the seed and pulp extracts of C. millenii reduced the bacterial load in kunu-zaki. Their finding is line with the report of Giwa et al., (2012) on the antimicrobial property of C. milleniseed and pulp.

Table 4. reveals the associated fungi organisms isolated from C. parchycarpa and their corresponding percentage incidence. Four fungi islates namely Fusarium moniliforme, F. oxysporum, Aspergillus niger and A. flavuswere isolated and they all proved to be pathogenic. A. flavus recorded the highest incidence (40%) and this was followed by F. moniliforme (30%) and F. oxysporum (20%) respectively. A. niger had the least percentage incidence of 10%. Little is known about the mycoflora ofC. parchycarpa fruit but earlier study has shown the presence of spoilage microorganisms in C. parchycarpa juice and jam (Okudu & Ene-Obong, 2015). However, the occurrence of the above isolatedspoilage fungi on other tropical fruits have been reported by early researchers, showing their pathogenic and deteriorative ability (Zacharia &Philip, 2010; Ahmed and Mohammed, 2014; Chuku, 2005 & 2012).

IV. Conclusion

C. parchycarpa fruit is rich in abundant nutrients and provides necessary mineral elements required for a healthy living. Improper handling predisposes this endowed fruit to organisms especially fungi that are capable of causing spoilage. Therefore, proper and hygenic measures should be followed in order to prevent contamination by these organisms with a sole aim of providing healthy fruits that are safe for consumption.

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