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Isolation and Identification of Fungi Associated with Dried Fruits of *Detarium microcarpum Guill. & Perr.* Fruits

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ABSTRACT

This study aimed at isolated and identified fungi associated with the dry fruits of Detorium microcarpum which fresh fruits were collected and dried at room temperature in Dutse Local Government of Jigawa State. The fruits of Detarium microcarpum were surface sterilized with cotton wool soaked in 70% alcohol. The pulps of the sample fruits were extracted with the help of pestle and mortar, one (1g) gram of the pulp was introduced into a beaker containing nine miles (9ml) of distilled water. A drop of the serially diluted solution was then plated on solidified Potato Dextrose Agar plates (90mm diameter) aseptically. Inoculated plates were incubated at 28±30^{0}C for 7days. A small portion of the aerial mycelia from the representative culture was picked using a sterile inoculating needle and inoculated on a slide for 24-48 hours, after which it was viewed under the light microscope to detect spore, hyphae and other special structures. The results shows that Rhizopus stolonifer, Apergillus niger Fusarium oxysporum, Fusarium moniliforme were found to be associated with the dry fruits of Detarium microcarpum with frequencies of occurrence of 42%, 32%, 10% and 16% respectively. The disease-causing potential of this two isolates Aspergillus niger Fusarium moniliforme, seems to be very serious in human as Aspergillus niger have been responsible for causing Pneumonia and several bronchial diseases while Fusarium moniliforme has champion the production different kinds of toxin that may have health implication to human and other animals. It is therefore recommended that, there is the need for proper storage of Detarium microcarpum fruits so as to preserve its nutritive value. Educational activities should be organized to local communities on the best practices of preserving the fruits of Non Timber Forest Product.

Keywords: *Detorium microcarpum, dry fruits, Fungi and Jigawa state*

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INTRODUCTION

In most developing countries, food shortage is becoming evident as a result of population growth, competition for fertile land and poverty. The diet of many rural and urban dwellers is deficient in protein and high in carbohydrates, the implication is high incidence of malnutrition and increased dietary disease a situation in which children and lactating women are most vulnerable (Awuchi *et al,* 2020). Thus, the pulp of this fruit is known to be rich in sugar, vitamin C and certain minerals (Eromosele *et al*., 1994). Dogara, (2022) state the fruit, bark and leaves of plants such as Sweet detar (*D. microcarpum*) are used not only for medicine, texture and flavor, but also for their nutritional values. The seed coat of the fruit has been shown to possess antimicrobial activity due to the presence of steroidal saponins and flavonoids (Ebi and Afieroho, 2011). *D. microcarpum* is a fruit tree of great socioeconomic importance in West Africa (Agbo, 2017). The seed contained in the stone of the fruit is also rich in nutritional and functional properties and has the particularity of being rich in hydro colloids (59.8g-100g^{-1}). Its hard dark-brown wood provides very good quality timber which is very durable under water and is used in carpentry and construction. It is also used as good quality fuel wood and charcoal. The leaves, stems, roots, barks, as well as the fruits have found tremendous usage in treatment of various ailments like tuberculosis, diabetes, itching and diarrhea (Makalao *et al.,* 2016). The fruit is rich in vitamin C and the leaves and seeds are also used in cooking. The fruit may be eaten raw or cooked, but traditionally, the mesocarp is transformed into flour used in the preparation of cakes, bread, couscous, baby food and local beer. Seed kernels are added to Egusi soup (generic name for seeds of some Cucurbitaceae species) or are cooked and eaten as a vegetable (Pancel, 1993).

World Health Organization reports that "wild" plants may support the health and nutritional needs of 80% of people living in developing countries (Van Andel, 2006). While every measure is being taken by various levels of government to boost food production by conventional agriculture, a lot of interest is currently being focused on the possibilities of exploiting the vast number of less familiar plant resources of the wild (Anwhange *et al.,* 2004; Abdullahi and Abdullahi, 2005). Non-timber forest products (NTFPs) would therefore represent a considerable food and economic stake for local populations. *Detarium macrocarpum* were identified among the Non Timber Forest Products (NTFPs) of major importance in Africa and thus, it is recognized that rational exploitation of local resources, through improved processing technologies, would make a valid contribution to the sustainable development of the continent (Kouyate and Lamien, 2011). Sweet detar, *Detarium microcarpum spp*. belongs to the class of Magnoliopsida, subclass of Rosidae, order of Fabales, family of Fabaceae, subfamily of Caesalpinioideae, tribe of Detarieae (Kouyate, 2005). *Detarium species* has several species including *Detarium microcarpum* (Akah, 2012). *Detarium microcarpum* inhabits dry lands, wooded Savannas, and open forests of the Sudano-Guinean and Sudano-Sahelian zones of the African continent (Arbonnier, 2002). Its range extends from Senegal to Sudan, in particular in Benin, Burkina Faso, Cameroon, Côte d'Ivoire, Gambia, Ghana, Guinea, Guinea Bissau, Mali, Niger, Nigeria, Central African Republic, Senegal, Sudan and Chad (Kouyate, 2005). *Detarium microcarpum* bears different local names among socio-cultural groups of different countries. For examples Yoruba, Igbo, Kanuri and Hausa in Nigeria named the plant as Ogbogbo, Ofo, Gatapo and Taura while Fulbe, Sonrai and Soninke in Mali called it Doli, Tambacounba and Fantu respectively (Kouyaté, 2005). It is known as: "sweet dattock" or "tallow tree" or "sweet Detar" in English and "Détar sucré" or "petit Détar" in French.

Fungi

Fungi are a distinct kingdom of organisms that are neither plants nor animals. The basic fungal vegetative structure is the microscopic hyphae, a thread like-tube that maybe separated into cells by the formation of cross-walls (septa). Unlike plants and animals' fungi absorb all their food from external sources. The hyphae grow through or on their food substrate and sometimes form a visible mycelium. Many fungi are saprophytic or parasitic (Orwa *et al*., 2009). There is considerable variation in the structure, size, and complexity of various fungal species. For example, fungi include the microscopic yeasts, the molds seen on contaminated bread, and the common mushrooms (Figures 1 and 2).

Figure 1: Detarium tree and fruits

Figure 2: Detarium fruits

Detarium microcarpum is a leguminous tree from West Africa that bears pods containing sweet sour pulp which is popularly eaten by local people. The fresh fruits has been found to have been eaten raw, thus the consumption of ineffectively preserved fruit of *Detarium microcarpum* may have health implication as they could be jam-packed of

fungi which could reduce the quality of the fruit and invariably leads to one form of ailment or another, therefore, this research intends to isolate and identify fungi associated with dry fruits of *Detarium microcarpum* so as to ascertain whether there are pathogenic strains among them.

MATERIALS AND METHODS

The study was carried out at Federal University Dutse, Jigawa State. Dutse is a city located in Northern Nigeria, with coordinates of latitude11^{0}42'04^{0}N and 9^{0}20'31^{0}E and longitude 11^{0}70'11^{0}N, 9.3^{0}41'94"E and elevation of 435m above the sea level. The rainy season lasts from May to September with an average of annual rainfall of between 600mm 1000mm and high temperature are normally recorded between the month of April and September 2009. The inhabitants are predominantly farmers engage in farming and rearing of livestock (Richard *et al*., 1997). Dutse is predominantly occupied by Hausa and Fulani with the estimate population of 153,000 (NBS, 2006). The topography is characterized by high land area which is almost 750m. Soil tends to be fertile ranging from sandy - loamy (Salami *et al*., 2020; Salami and Lawal, 2018).

Sample collection

Fresh fruits of *Detarium macrocarpum* were collected from the mother tree of the species from Jigawar Sarki, Kudai and Gidan makera of Dutse Local Government in Jigawa State in December. They were allowed to dry at room temperature for about three (3) months. All the sampled fruits were mixed thoroughly to ensure each fruit had equal chance of being used in the experiment.

Sample Preparation and inoculation

The pulps of the sample fruits were extracted with the help of pestle and mortar, one (1g) gram of the pulp was introduced into a beaker containing nine miles (9ml) of distilled water. The sample solution was serially diluted in this manner 10^1, 10^2, 10^6 in order to reduce fungal load in the sample. A drop of the serially diluted solution was then plated on solidified Potato Dextrose Agar plates (90mm diameter) aseptically. The inoculated plates were incubated at 28±30^{0}C for 7days. From the incubated plates the different fungal isolates with different colorations observed includes; (i) Brown (ii) Black (iii) Green and (iv) White which signified the occurrence of different fungal colonies. The fungal colonies that emerged were continuously sub-cultured in order to obtain a pure culture of the fungal isolates.

Identification of the Fungal Isolates

The one to four weeks pure cultures of the fungal I solates were identified using cultural and morphological features such as colony growth pattern, conidial morphology and pigmentation (Oyeleke and Manga, 2008), by slide culture techniques (Abdullah *et al*., 2016). A small portion of the aerial mycelia from the representative culture was picked using a sterile inoculating needle and inoculated on a slide containing a fraction of a prepared solidified Potato Dextrose agar and incubated for 24-48 hours, after which it was viewed under the light microscope first with (x10) and then with (x40) objective lens to detect spore, hyphae and other special structures. The Morphological characteristics and appearance of the fungal isolated from the dry fruits of *Detarium microcarpum* used in this study were confirmed and authenticated with the help of Microbiology manual by Cappuccino and Sherman (2014).

RESULTS

The fungal isolation results indicate that *Rhizopus stolonifera* had the highest prevalence among the identified organisms, accounting for 42% of the total colonies, suggesting its dominance in the sampled environment. *Aspergillus niger* followed closely with 32%, reflecting its adaptability and common presence in various ecological niches. The two Fusarium species were less frequently encountered, with *Fusarium moniliforme* constituting 16% and *Fusarium oxysporum* 10%, respectively. Although these Fusarium species appeared less frequently, their presence is significant as they are known plant pathogens and may pose risks in agricultural or stored-product settings. The fungal profile reveals a diverse community, with Rhizopus and Aspergillus being the most prominent (Table 1).

Table 1: Fungal colonies identified in Dry *Detarium microcarpum* fruits in percentages

Organism	Colonies identified	Percentage (%)
Aspergillus niger	6	32
Fusarium moniliforme	3	16
Fusarium oxysporum	2	10
Rhizopus stolonifera	8	42

DISCUSSION

The isolation and identification of fungi associated with dried fruits of *Detarium microcarpum* (Guill. & Perr.) have significant implications for food safety, public health, and postharvest management (Dogara, 2022). Fungi such as *Aspergillus niger, Fusarium spp.,* and *Rhizopus stolonifera* are known to cause spoilage and reduce the nutritional and economic value of stored fruits (Adeniyi *et al.,* 2024; Liu et *al.,* 2024). More critically, some species, especially from the Aspergillus and Fusarium genera, are capable of

producing mycotoxins like aflatoxins and fumonisins, which pose serious health risks to consumers, including liver damage and carcinogenic effects. Identifying these fungal contaminants helps in developing better storage, drying, and packaging techniques to reduce fungal growth, improve shelf-life, and ensure the safety of *Detarium microcarpum,* which is an important wild fruit with nutritional and medicinal value in many African communities.

Results shows that *Rhizopus stolonifer, Apergillus niger Fusarium oxysporum, Fusarium moniliforme* are found to be associated with the dry fruits of *Detarium microcarpum* with frequencies of occurrence of 42%, 32%, 10% and 16% respectively. This is in agreement with that of Ugwu *et al*., (2014) which discovered six fungal species namely *Candida tropicalis, Penicillium notatum, Aspergillus niger, Fusarium oxysporum, Absidia corynbifera, Rhizopus stonolifer* and four species of bacteria namely *Escherichia coli, Klebsiella spp., Salmonella spp.* and *Pseudomonas* that cause post-harvest spoilage in Pepper fruits. Moreover, Kafinta *et al.,* (2013) identified *Apergillus fumigatus, Apergillus niger, Aspergillus flavus* and *Rhizopus stolonifer* to be associated with the spoilt sweet orange fruit *(Citrus sinens*is)

Several studies have also reported that *Aspergillus spp* is associated with spoilage of plants fruits, seeds, and fruits such as apricot, orange, lemon, peach, apple, kiwi, mango Chukwuka, *et al*., (2010) reported that *Aspergillus* had the highest decay diameter among other fungi associated with *Detarium microcarpum* plant spoilage. The occurrence of *Aspergillus* in rotten *Detarium microcarpum* seed could pose a serious health risk, especially when the plant is not well prepared. Some molds are capable of producing more than one mycotoxin and some mycotoxins are produced by more than one fungal species. Marasas *et al* (1984) states, Toxins produced by *F. moniliforme* are fusaric acid, fusarins, gibberellins, moniliformin, and fumonisins.

These fungi are not only known agents of spoilage but are also capable of producing harmful mycotoxins such as aflatoxins and fumonisins, which have been linked to liver cancer, immune suppression, and other chronic health conditions. Recent studies in Nigeria confirm the widespread presence of these fungi in dried food products. For instance, Esan *et al.* (2023) found mycotoxin contamination in ready-to-eat foods due to Aspergillus and Fusarium species, while Abdul-Rahman *et al.* (2023) reported similar findings in dried tomato chips. These results underscore the need for improved postharvest practices, including better drying, packaging, and storage techniques, as well as community education to prevent health risks and ensure the safe use of *Detarium microcarpum,* which holds both nutritional and medicinal value in local communities. Addressing fungal contamination is also crucial to preserving the economic potential of this underutilized wild fruit in rural African livelihoods.

Conclusion

The presence of fungi such as *Rhizopus stolonifer, Aspergillus niger, Fusarium moniliforme*, and *Fusarium oxysporum* in dried *Detarium microcarpum* fruits confirms that postharvest fungal contamination is a significant threat to the safety and quality of this wild edible fruit. The high frequency of Rhizopus (42%) and Aspergillus (32%) suggests these fungi are major contributors to spoilage, with potential implications for public health due to their ability to produce harmful mycotoxins. These findings align with previous studies reporting similar fungal species in the spoilage of various fruits. Therefore, effective postharvest handling, storage, and processing methods are essential to reduce contamination risks, preserve nutritional value, and prevent possible toxin exposure from consumption.

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