



Research Article

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Effects of Untreated Cassava Mill Effluent on the Mycological Properties of Soil in Aba, Abia State, Nigeria

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Abstract

Cassava is a major staple food in the developing world, providing a basic diet for over half a billion people. Nigeria is the world's largest producer of cassava, while Thailand is the largest exporter of cassava starch. Two important wastes are generated during processing of cassava tubers. These are cassava peels and liquid squeezed out of mash. Cassava effluent, is a pale-yellow turbid liquid with an earthy but offensive odour. Waste water from cassava processing is odorous and its microbial content is often high. The indiscriminate discharge of cassava can lead to the pollution of soil. The impacts of cassava effluent are hazardous to human health, animal lives, plant lives and the economy. This has led to this work which aimed at evaluating the effects of untreated cassava mill effluent on the mycological properties of soil in Aba, Abia State, Nigeria. Fifteen samples which comprise of five samples each from fresh cassava effluent sample, cassava effluent-contaminated soil sample and uncontaminated soil samples were collected from five different locations in Aba and were serially-diluted. Total fungal count was determine using the spread place technique. The mycological load of the fresh effluent (5×10^6 - 8×10^6 cfu/g) and effluent-contaminated soil (3×10^6 - 6×10^6 cfu/g) was lower than the $x1=6-9 \times 10^6$ cfu/g) uncontaminated soil sample. The relatively low population of fungus in the fresh effluent and effluent contaminated soil could be attributed to the acidic nature of the effluent due to the presence of cyanide. Five mould species and one yeast were isolated from the samples. They were *Aspergillus niger*, *Aspergillus fumigatus*, *Rhizopus arrhizus*, *Fusarium solani*, *Mucor heimalis* and *Sacchromyces cerevisiae*. Eight, Seventeen, and Nineteen of the untreated effluent, effluent-polluted soil and unpolluted soil had the fungal isolates. The unpolluted soil supported the growth of the isolates except *Mucor hiemalis*. *Aspergillus niger* occurred most frequently in the unpolluted soil (28%). The untreated effluent adversely affected the fungal populations which might have serious consequences in their ecological role, therefore adequate treatment of the effluent before discharge into the soil environment is crucial.

Introduction

Soil is a major component of the earth's ecosystem [26]. It acts as an engineering medium, a habitat for soil organisms, a recycling system for nutrients and organic wastes, a regulator of water quality,

and a medium for plant growth, making it a critically-important provider of ecosystem services [31]. *Manihot esculenta*, commonly called cassava USDA, (2014), is a woody shrub of the spurge family,



Euphorbiaceae, native to South America, from Brazil and parts of the Andes. Although a perennial plant, cassava is extensively cultivated as an annual crop in tropical and subtropical regions for its edible starchy root tuber, a major source of carbohydrates. Cassava is predominantly consumed in boiled form, but substantial quantities are used to extract cassava starch, called tapioca, which is used for food, animal feed, and industrial purposes. Cassava is the third-largest source of food carbohydrates in the tropics, after rice and maize *FAO* (2011); *Claude and Denis* (1990); *Lominda* (2019). It is a major staple food in the developing world, providing a basic diet for over half a billion people *FAO* (1995). It is one of the most drought-tolerant crops, capable of growing on marginal soils. Nigeria is the world's largest producer of cassava, while Thailand is the largest exporter of cassava starch. It is classified as either sweet or bitter. Like other roots and tubers, both bitter and sweet varieties of cassava contain anti-nutritional factors and toxins, with the bitter varieties containing much larger amounts *FAO* (1990).

Oboh (2005) [27] identified two important wastes that are generated during processing of cassava tubers to include cassava peels and the liquid squeezed out of the mash. Cassava effluent, is a pale-yellow turbid liquid with an earthy but offensive odour. It contains large floating or suspended solids and very small solids in colloidal suspension [2]. The cassava effluent may also contain oil and grease from the lubricated parts of the grinding machine, in addition to its normal composition of carbohydrates and organic solids. Its processing is generally considered to contribute significantly to environmental pollution and aesthetic nuisance. The waste water also contains heavy loads of microorganisms, lactic acid, lysine (from *L. coryneformis*) and amylase (from *L. delbruckii*) capable of hydrolyzing the glycosides [32,2]. Waste water from cassava processing is subjected to relatively rapid breakdown. It is odorous and its microbial content is often high. There is the additional problem of the presence of simple and complex cyanide with degree of solubility, toxicity and stability [11]. One of the major threats to the environment by garri processing industry is the hydrocyanic acid and the unbroken down cyanogenic glycoside-linamarin and lotaustralin which produce toxic and acidic effects, to the soil, soil organisms, water and plants. Other by products such as uncoverable starch, the peel and waste water could result in the generation of objectionable odor and constitute breeding ground for flies and insects, which are carriers of diseases. The peel not only contain cyanogenic glycosides and free cyanide which can be degraded and leached into the soil. The waste water could also change some other soil properties because of its total solids, total organic carbon, nitrogen and phosphorus [3].

The indiscriminate discharge of cassava can lead to the pollution of soil. Cassava effluent discharged into the land and the subsequent washing down of the pollutants to successive horizons. These effluents discharged into the land contain many toxic chemicals, mineral acids, bases, etc. which over the period of time get deposited into the soil due to their retention or their absorption in

the soil particles. This may affect the growth of fungal and bacterial colony in the soil. Some of the deposited chemicals may also be taken up by the plant/crops growing in such contaminated soil [3]. Fungi perform important functions within the soil in relation to nutrient cycling, disease suppression and water dynamics, all of which help plants become healthier and more vigorous [22]. Along with bacteria, fungi are important decomposers of hard to digest organic matter. They use nitrogen in the soil to decompose woody carbon-rich residues low in nitrogen and convert the nutrients in the residues to forms that are more accessible for other organisms [22]. The aim of this work is therefore to evaluate the effects of untreated cassava mill effluents on the mycological properties of soil Abia, Abia State, Nigeria.

Materials and Methods

Collection of Samples

Fresh cassava mill effluents were collected from five different cassava mill dumps in Iheorji, Iheorji avenue, Umuogele, 16 Dike Street and, Owerri Aba in Aba, Abia State Nigeria using sterile syringes. These were collected into sterile 1-liter rubber containers with screw caps. Cassava 15cm effluent-contaminated soil were collected from a depth of 15cm using a sterilized soil auger into sterile screw cap containers while the uncontaminated soil (control) was collected from a site 100m away from the milling dumps. A total of fifteen effluent samples were collected. All containers used for mycological analysis were sterilized with 70% ethanol and then rinsed with sterile distilled water and thereafter with the effluent water. The samples were transported in an ice-packed box and analyzed within 24 hours of collection at the microbiology Laboratory of Abia State Polytechnic Aba, Abia State, Nigeria.

Sample Preparation

The cassava effluents were sieved through a 2mm stainless steel sieve to remove the debris and stored in 1 liter-plastic containers, while the soil samples were subjected to air drying for a period of one week in a clean well-ventilated laboratory, homogenized by grinding, passed through a 2mm stainless sieve and stored in labeled plastic containers until analysed.

Serial Dilution and Inoculation

Ten folds serial dilutions of the samples were prepared as described by *Cheesbrough* (2006) [7]. One milliliter of the effluent and one gram of the polluted soil and unpolluted soil were inoculated each into 9ml sterile water. One ml of the effluent and homogenized soil samples was transferred into the second tube containing 9ml sterile water. This was continuously repeated until the tenth tube (10-10). Diluted samples were used for the mycological analysis.

Inoculation and Enumeration of Fungi

The total fungal count was determined using spread plate technique as described by *Cheesbrough* (2006) [7]. Sabouraud

dextrose agar was prepared based on the manufacturer's instruction. Chloramphenicol was introduced into the agar to inhibit bacterial growth. 0.1ml of the serially-diluted samples (10⁻⁶) was dispensed into the Centre of a Sabouraud dextrose agar medium plate and spread evenly over the surface. Incubation was carried out in an inverted position at room temperature for 72 hours after which the fungal colonies that developed were counted. The result was recorded and expressed as colony forming unit per milliliter of the effluent sample (cfu/ml) and per gram for the soil samples (cfu/g) respectively. Each colony was sub-cultured and stored in a sterile Sabouraud dextrose agar slant for characterization and identification.

Characterization and Identification of the Fungal Isolates

Colony morphology, cellular morphology, Germ tube test, Slide culture, Gram Staining, Carbohydrate assimilation, Urease and motility tests were carried out to characterize and identify the fungi as described by *Cheesbrough* (2006) [7]. They were identified as to the description of *David et al.* (2007) [8].

Colony Morphology and Cellular Morphology

The colony morphology determination was based on the color, elevation, margin and shape while the cellular morphology determination was based on the shape and arrangement of the cells.

Germ Tube Test

This test was performed as described by *Cheesbrough* (2006) [7]. 0.5ml of human serum was dispensed into a test tube. The serum was inoculated with yeast colony. The test tube was incubated at 37 °C for 3 hours. A Pasteur pipette was used to transfer a drop of the serum yeast culture to a clean slide and covered with a cover slip. It was examined using 10x and 40x objectives. Sprouting yeast cell in the form of a tube-like out growth from the cell were recorded as a positive result.

Slide Culture Test

The method described by *Cheesbrough* (2006) [7] was adopted. Sterile Sabouraud dextrose agar medium was inoculated with a mold isolate. A cover slip was be specific inserted beside the mold. It was incubated in an inverted position for 4-7 days. The cover slip was placed on a slide containing a drop of lactophenol blue and was examined with 10x and 40x microscope objectives. The microscopic appearance was recorded.

Gram Staining

Gram staining was carried out as described by *Cheesbrough* (2006) [7] to determine the gram reaction of the yeast isolate. A smear of the yeast isolates from each of the sample of the effluent, polluted and unpolluted soil was made on a clean slide and allowed

to dry. It was heating fixed by passing the smear through the Bunsen burner. This was done to enhance the sticking of the organism to the microscope slide. The smear was flooded with crystal violet and left for 60 seconds before washing off clean water. Lugol's iodine was added and allowed to stand for 60 seconds before being washed off and decolorized with ethanol for 10 seconds. The slide was then washed off, stained with safranin for two minutes, washed off and allowed to air-dry. A drop of immersion oil was added to the slide which was then viewed under the microscope using 100x objective lens.

Carbohydrate Assimilation Test

This test is used to detect organisms which utilize different sugars as source of energy with the production of acid and/or gas. The sugars used were glucose, sucrose, lactose and mannitol. Peptone water broth was prepared based on manufacturer's instruction. Bromothymol blue indicator was added to the broth. In four separate conical flasks containing glucose, sucrose, lactose and mannitol, the above solution (peptone water broth + indicator) was added at equal proportions. Five milliliters of the mixture were then dispensed into test tubes. Durham tubes were added in an inverted position and the test tubes were sterilized in an autoclave at 121°C for 15 minutes. A change in color of the mixture indicated acid production while gas production was indicated by a void in the Durham tubes.

Motility Test

This was carried out as described by *Cheesbrough* (2006) [7]. Each yeast isolate was separately inoculated into a semi-solid nutrient agar medium using a sterile straight wire and incubated at 37 °C for 24 hours. Migration of the isolates away from the line of inoculation was recorded as a positive result while lack of migration away from the line of inoculation indicated a negative result.

Urease test

This was carried out as described by *Cheesbrough* (2006) [7]. The medium used was Urea broth. This test is used in differentiating the Enterobacteria. The test tubes were sterilized and 2ml of the broth was dispensed into the test tubes. The test organism was heavily inoculated into the test tube and incubated at room temperature for 48 hours. The development of a pink color indicated a positive result.

Data Analysis

The data collected were subjected to statistical analysis using IBM SPSS version 23. One-way analysis of variance was used to show significant difference at $P < 0.05$.

Results

The average fungal counts of the untreated cassava mill effluent; untreated cassava mill effluent-polluted soil and unpolluted soil are

presented in Table 1. The counts ranged between 5×10^6 cfu/ml, and 8×10^6 cfu/ml, 3×10^6 cfu/mg and 6×10^6 cfu/mg, and 6×10^6 cfu/mg and 9×10^6 cfu/mg for untreated cassava mill effluent, untreated cassava mill effluent-polluted soil and unpolluted soil respectively. The untreated cassava mills effluent sample from Umuogele and Owerri Aba had the highest count while 16 Dike Street had the least

court. Untreated cassava mills effluent-polluted soil sample from Owerri Aba had the highest count while 16 Dike had the lowest count. Unpolluted soil sample from Iheorji Avenue Street and 16 Dike Street had the highest count while Iheorji had the least count (Table 1).

Table 1: Average fungal counts untreated cassava mill effluent, untreated cassava mill effluent-polluted soil and unpolluted soil.

Sample	Cassava Mill Location	Untreated Cassava Mill Effluent (cfu/ml)	Untreated Cassava Mill Effluent Polluted Soil (cfu/g)	Unpolluted Soil (cfu/g)
1	Iheorji	7×10^6	5×10^6	6×10^6
2	Iheorji Avenue	6×10^6	4×10^6	9×10^6
3	Umuogele	8×10^6	4×10^6	7×10^6
4	16 Dike	5×10^6	3×10^6	9×10^6
5	Owerri Aba	8×10^6	6×10^6	7×10^6

Table 2 showed the colonial and microscopic characteristics of the moulds isolated from cassava mill effluent, effluent-polluted soil and unpolluted soil. The isolates were *Aspergillus niger*, *Aspergillus*

fumigatus, *Rhizopus arrhizus*, *Fusarium solani* and *Mucor heimalis* (Table 2 and Table 3).

Table 2: Colonial and microscopic characteristics of the mould isolated from cassava mill effluent, effluent-polluted and unpolluted soil.

Isolates	Colonial Characteristics	Microscopic Characteristics	Organisms
1	Colonial consisted of a compact white or yellow basal felt covered by a dense layer of dark brown to black conidial heads	Conidial heads were large, globose, dark brown, becoming radiate and tending to split into several loose column with age. Conidiophore stipes were smooth-walled hyaline or turning dark towards the vesicle. Conidial heads were biserate with the phialides borne on brown, often septate metuluate. Conidial were globose to subglobose, dark brown to black and rough-walled	<i>Aspergillus niger</i>
2	Colonies were typically blue-green with suede-like surface consisting of dense felt conidiophores	Conidial heads were typically columnar and uniseriate. conidiophores stipes were short smooth walled and had a conical shaped terminal vesicle which support a single row of phialides on the upper two thirds of the vesicles. Conidia were produced in basipetal succession forming long chains and were globose to subglobose, green and finely roughened	<i>Aspergillus fumigatus</i>
3	Colonies were very fast growing with some tendency to collapse, white cottony at first becoming brownish grey to blackish-grey depending on the amount of sporulation	<i>Sporangiospores</i> were smooth-walled, non-septate, simple or branched, arising from stolons opposite rhizoids usually in groups of three or more. sporangia were globose, with a flattened base grayish black, powdery in appearance and mainly spored. columnellae and apophysis together were globose, subglobose or oval, collapsing to an umbrella-like form after spore release. sporangiospores were angular, subglobose to ellipsoidal, with striations on the surface	<i>Rhizopus arrhizus</i>
4	Colonies were rapidly growing, aerial mycelium white to cream, becoming bluish-brown when sporodochia were present. Macroconidia were from short multiple branched conidiophores which may form sporodochia. they were three to five five-septate, cylindrical, often moderately curved, with an indistinct pedicellate foot cell and a short blunt apical cell	Microconidia were usually abundant, cylindrical to oval and formed from long lateral phialide. Chlamydospores were hyaline, globose, smooth to rough-walled, in pairs on short lateral hyphal branches	<i>Fusarium solani</i>
5	Colonies were typically grey	Branched sporangiophores that yielded yellow to dark brown sporangia which can mate to form black-brown, spiny zygosporangia were seen.	<i>Mucor heimalis</i>

Table 3: Morphological and biochemical characteristics of yeast isolate from cassava mill effluent, effluent-polluted and unpolluted soil.

Isolate	Colour	Shape	Urease	Germ tube	Glucose Assimilation	Lactose Assimilation	Mannitol Assimilation	Sucrose Assimilation	Motility	Organism
1	Cream	ellipsoidal	-	-	+	+	+	+	-	<i>Saccharomyces cerevisiae</i>

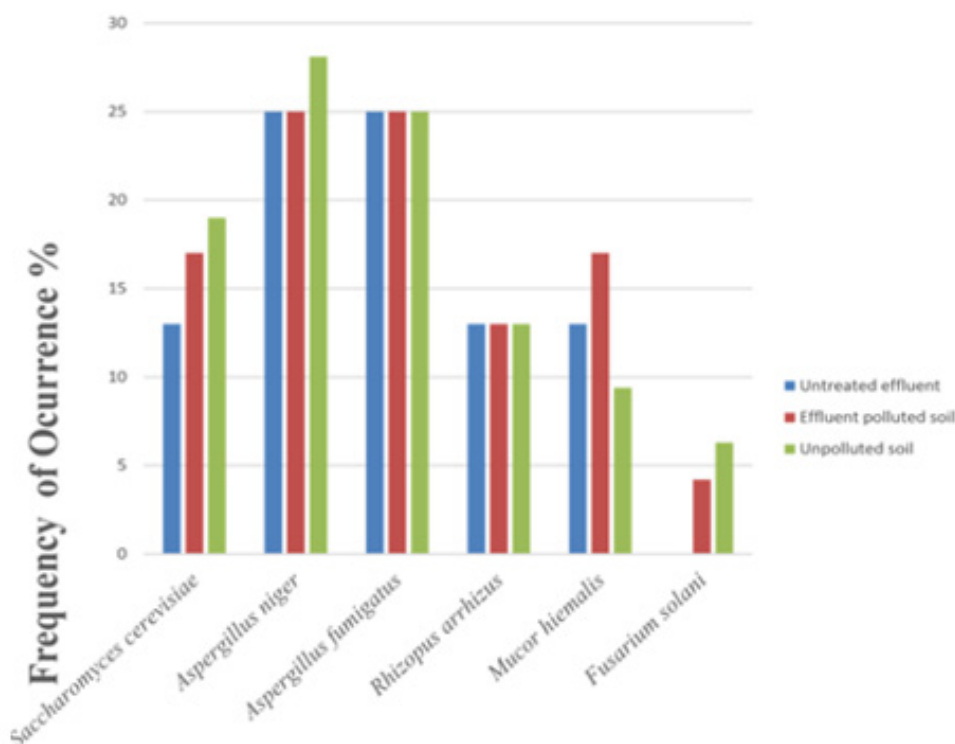
Morphological and biochemical characteristics of yeast isolate from cassava mill effluent; effluent-polluted and unpolluted soil are shown in table 3. The yeast isolated was *Saccharomyces cerevisiae*. The number of untreated cassavas mill effluent, untreated cassava mills effluent-polluted soil and unpolluted soil with fungal isolate as shown in table 4. The untreated cassava mill effluent contained *Aspergillus niger* 4(80.0), *Aspergillus fumigatus* 4(80.0), *Rhizopus arrhizus* 3(60.0), *Fusarium solani* 1(20.0), *Mucor heimalis* 2(40.0) and *Saccharomyces cerevisiae* 3(60.0) respectively. The unpolluted soil contained *Aspergillus niger* 5(100.0), *Aspergillus fumigatus*

4(80.0), *Rhizopus arrhizus* 3(60.0), *Fusarium solani* 2(40.0), *Mucor heimalis* 2(40.0) and *Saccharomyces cerevisiae* 3(60.0) respectively (Table 4).

The frequency of occurrence of the fungal isolates in the untreated effluent-polluted soil and unpolluted soil are presented in Figure 1. *Aspergillus niger* (25%) and *Aspergillus fumigatus* (25%) occurred most frequency in the untreated effluent, polluted and unpolluted soil while *Aspergillus niger* (28%) occurred most frequency in the unpolluted soil (Figure 1).

Table 4: Number of untreated cassava mill effluent, untreated cassava mill effluent-polluted soil and unpolluted soil.

Fungal Isolate	Untreated Cassava Mill Effluent (%)	Untreated Cassava Mill Effluent-Polluted Soil (%)	Unpolluted Soil (%)
<i>Aspergillus niger</i>	1 (20.0)	4 (80.0)	5 (100.0)
<i>Aspergillus fumigatus</i>	1 (20.0)	4 (80.0)	4 (80.0)
<i>Rhizopus arrhizus</i>	1 (20.0)	3 (60.0)	3 (60.0)
<i>Fusarium solani</i>	0 (0.0)	1 (20.0)	2 (40.0)
<i>Mucor heimalis</i>	2 (40.0)	2 (40.0)	2 (40.0)
<i>Saccharomyces cerevisiae</i>	3 (60.0)	3 (60.0)	3 (60.0)

**Figure 1:** Frequency of isolation of fungi in the untreated effluent, untreated effluent polluted soil and unpolluted soil.

Discussion

The average total fungal count of the untreated cassava mill effluent ranged from 5×10^6 – 8×10^6 cfu/ml while that of the untreated cassava mill effluent-polluted soil ranged from 3×10^6 – 6×10^6 cfu/g. The unpolluted soil had a total fungal count that ranged from 6×10^6 – 9×10^6 cfu/g Table 1. These high fungal counts may be due to lack of efficient control measures in the discharge of the waste water into the environment [4]. Uzochukwu, et al., (2001) [37] reported that high level of cassava wastewater is produced daily and drained into roads, streets, rivers and agricultural lands in garri processing communities in Nigeria. These singular activities tend to expose the wastewater to microbial contamination.

The mould species isolated were *Aspergillus niger*, *Aspergillus fumigatus*, *Rhizopus arrhizus*, *Fusarium solani* and *Mucor heimalis* Table 2 while the yeast, was *Saccharomyces cerevisiae* Table 3. The unpolluted soil enabled the growth of more fungal isolates than the effluent-polluted soil and untreated effluent. This could be attributed to the less acidic nature of the unpolluted soil.

Aspergillus niger occurred in 1 (20%), 4 (80%) and 5 (100.0%) of the untreated effluent, untreated effluent polluted soil and unpolluted soil respectively, *Aspergillus fumigatus* 1 (20%), 4 (80%), 4 (80%); *Rhizopus arrhizus*, 1 (20.0%), 3 (60.0%), 3 (60.0%); *Fusarium solani*, 0 (0.0%), 1 (20.0%), 2 (40.0%); and *Mucor heimalis*, 2 (40.0%), 2 (40.0%), 2 (40.0%); *Saccharomyces cerevisiae* occurred in 3 (60.0%), 3 (60.0%), 3 (60.0%) of the untreated effluent, untreated effluent polluted soil and unpolluted soil respectively Table 4.

The microbial population of the unpolluted soil was higher than that of the other samples. The relatively lower population of fungus in the effluent-polluted soil could be attributed to the acidic nature of the effluent due to the presence of cyanide. Cyanide in the soil and fermented cassava could lead to the inhibition of microbial growth [10]. Disposal of cassava wastes from processing activities in mills lead to the release of a wide variety of microorganisms. These organisms may release toxins in the effluent which can be harmful [12,13]. Only those that can withstand the high acidic condition of the processing wastes will dominate, thus the lower population of the fungal species. The absence of the acidic effluent in the unpolluted soil is attributable to the 100% occurrence of the isolated organisms.

These findings indicated that the untreated effluent, untreated effluent-polluted soil and unpolluted soil promoted the proliferation of *Aspergillus niger* and *Aspergillus fumigatus*. *Aspergillus niger* and *Aspergillus fumigatus* were more prominent in the unpolluted soil indicating that cassava mill effluent did not influence the proliferation of the other fungi species. *Aspergillus fumigatus* and *Aspergillus niger* were predominant in both the polluted and unpolluted soil (Figure 1). This is an indication that the cassava mill

effluent favored the growth of the fungus which could be due to a higher quantity of decaying organic matter in the polluted soil and unpolluted soil occasioned by the deposition of cassava effluent into the soil [29].

Saccharomyces cerevisiae is one of the main species of yeast that is widely utilized in several industrial sectors [20,19]. The choice of *Saccharomyces cerevisiae* in an industrial setting could be due to their ability to breakdown sugars to form ethanol and carbon dioxide [34]. *Saccharomyces cerevisiae* has been widely used for fermentation of different substrates. It is an essential microorganism that has been considered useful in animal feed to improve production and health status Suarez and Guevara (2018). *Saccharomyces cerevisiae* which are used as probiotics are nutritionally high in enzymes, fatty acids, vitamin B complex, unknown growth factors and amino acids (more than 40% of total dry matter) [34]. The organisms cultured in cassava mill effluents have been reported to contain essential heavy metals, low cyanide and cations and amino acids [19]. Amino acids, heavy metals, cations, etc have been reported in brewer's yeast [30]. Studies have shown that *Saccharomyces cerevisiae* contains crude protein, lipid, fibre and carbohydrate. Typically, cassava is known as a carbohydrate crop with low protein content, but during fermentation of carbohydrate substrate with *Saccharomyces cerevisiae*, the protein content is enhanced. Onyeulor and Nwaehiri (2018), reported that *Saccharomyces cerevisiae* (BY 4743) could enhance the protein content of potato peels through fermentation.

Aruna, et al., (2017) [5], reported protein enrichment of yam peels by fermentation using *Saccharomyces cerevisiae* (BY4743). Izah, et al., (2017) [19] reported that *Saccharomyces cerevisiae* could reduce the content of cassava mill effluents by 37.62%, 22.96%, 29.63%, 20.49%, 21.44%, 1.70%, 53.48%, 68.00%, 100%, and 74.48% for pH, conductivity, dissolved oxygen, total dissolved solid, salinity, sulphate, nitrate, phosphate, and chemical oxygen demand levels respectively, and increase turbidity by 17.17%. Izah, et al., (2017) [19], also reported that *Saccharomyces cerevisiae* has the tendency to uptake heavy metals in cassava mill effluents through biosorption, and reduce the concentration of cyanide in cassava mill effluents and the associated biomass. Previous studies have indicated that the organism improves the physical and chemical characteristics of cassava mill effluents [19]. Abioye, et al., (2015) [1] reported that *Saccharomyces cerevisiae* aid in the degradation of pharmaceutical effluents. Okoduwa, et al., (2017) [28], also reported the potentials of *Saccharomyces cerevisiae* in the treatment of tannery effluents. The nutrient uptake from the effluents lead to the possible increase in population of the *Saccharomyces cerevisiae*.

Aspergillus niger is commonly found in the soil, living as a saprophyte on decaying vegetation and leaves, compost piles, and stored grain. It is also found in indoor environments and as a contaminant of food. As the fungus is opportunistic, humans, birds, and animals can also be hosts, although this is extremely rare [33]. This fungus is spread via the air, soil, and water. It is commonly seen as a post-harvest disease due to its saprophytic nature. In the case

of humans and animals, a compromised immune system is typically present when the disease manifests. In plants, irrigation practices such as drip irrigation lines buried in soil and hot, humid growth conditions are conducive to disease development [33]. Many strains are used in commercial food production, including fermentation processes and the production of citric and gluconic acid [33]. It still causes several diseases including black mold (or black rot) of onions and garlic, crown rot of peanuts, and vine canker of table grapes. In black rot of onions and garlic, *A. niger* develops between the outer (dead, flaky) skin and the first fleshy scales of the bulb which become water soaked. The area dries and shrivels if the condition is dry, revealing black spore masses between outer scales. Other diseases attributed to *A. niger* include: tuber rot of yams, stem rot of *Dracaena*, black mold rot of cherry, kernel rot of maize, fruit rot of grapes, fruit rot of banana, rot of tomatoes, boll rot of cotton, root stalk rot of *Sansevieria*, and mango rotting [33]. Post-harvest decay results in discoloration, quality deterioration, and reduction in the commercial value of various crops [33]. In humans, rare cases have linked *A. niger* to Aspergillosis, a group of diseases that relate to the growth of and allergic responses to the fungus [33].

Aspergillus fumigatus is the most common and life-threatening airborne opportunistic fungal pathogen, which is particularly important among immunocompromised hosts [9,31]. Inhaling *A. fumigatus* spores (conidia) into the lungs may cause multiple diseases, which depend on the immunological status of the host in humans. These diseases include invasive pulmonary aspergillosis, aspergilloma, and different forms of hypersensitivity diseases such as allergic asthma, hypersensitivity, pneumonitis, and Allergic Bronchopulmonary Studied Aspergillosis (ABPA) [25,15]. Its presence in the samples studied could be due to the fact that they survive and grow on organic debris. *Aspergillus sp.* is known for aflatoxin production that destroys the liver by inducing fatty acid metamorphosis in cells [36].

Rhizopus arrhizus is a filamentous fungus that is the most common cause of mucormycosis. It is commonly found in dead organic matter. It is an opportunistic pathogen that causes human diseases in immunocompromised people, such as those with diabetes mellitus, cancer, or AIDS, by infecting pulmonary, cutaneous, and gastrointestinal tract, and facial sinuses, which expand to the brain. The symptom of these diseases is facial and eye pain, allergic reaction, bulging eyes, fever, cough and breathing difficulty. This microbe is very closely connected to human life [18]. Its presence in the polluted and unpolluted soil could be due to the fact that it is commonly found in dead organic matter which decomposes in the soil.

Mucor is a filamentous fungus found in soil, plants, and decaying fruits. The fungi cause the group of infections referred to as zygomycosis [21]. *Mucor hiemalis*, a common soil fungus of phylum *zygomycota*, is well known for producing a diverse array of enzymes and lipids Wang, (2007); Leck, (2009). Its presence in the sample maybe the ubiquitous nature of its spores as reported

by O'Gorman, *et al.*, (2009). Studies have shown that this mould produce mycotoxin which is known to be teratogenic, mutagenic, hepatotoxic, genotoxic and hepato carcinogenic depending on how long an individual gets exposed to it [14].

Fusarium solani is one of the most frequently isolated fungi from soil and plant debris and is also associated with serious invasive mycoses in immunocompromised and immunosuppressed patients [6,35]. They are ubiquitous in soil and decaying plant material, where they act as decomposers, but they are also host-specific pathogens of a number of agriculturally-important plants, including pea, cucurbits, and sweet potato. Moreover, they are increasingly associated with opportunistic infections of humans and other animals, causing systemic infections with a high mortality rate [23], as well as localized infections in the skin and other body parts [16,17]. Neutropenic patients, a category of particularly strongly immunocompromised patients, are susceptible to dissemination of infection from superficial or subcutaneous initiations. The infections are usually fatal [16,17,24].

Its presence in the effluent could be as a result of the methods of processing, milling and storage. Bacteria and fungi shedding from humans, pests and domesticated animals could have contributed to the elevated concentration of the organism in the unpolluted soil.

Saccharomyces cerevisiae, *Aspergillus niger*, *Aspergillus fumigatus*, *Rhizopus arrhizus*, and *Fusarium solani* were prominent in the unpolluted soil while *Mucor hiemalis* has the lowest growth in the unpolluted soil Figure 1. This is an indication that the cassava mill effluent did not form the growth of most of the fungi.

Conclusion

Disposal of cassava wastes from processing activities in mills lead to the release of a wide variety of microorganisms. Only those that can withstand the high acidic condition of the processing wastes will dominate, thus the lower population of the fungal species in the fresh effluent and effluent-contaminated soil. The fungal population of unpolluted soil was higher than the other samples. Most of the cassava millers in Aba are not educated and are not aware of the health and environmental effects of these wastes. Education on ways of detoxifying the wastes and proper disposal methods is therefore necessary so as to produce an environmentally-safe treated effluent suitable for disposal or reuse.

Conflict of Interest

None.

Acknowledgment

None.

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