

Original Research Article

FUNGAL and PHYSICOCHEMICAL CHARACTERISATION of WASTEWATER TREATMENT using CHARCOAL and CORN HUSK as FILTER MATERIALS.

ABSTRACT

Wastewater treatment is essential for managing water pollution, protecting public health, and maintaining a sustainable environment. It involves removing contaminants such as organic matter, chemicals, and pathogens from contaminated water before it is returned to natural water bodies or reused. This study investigates the comparative study of the fungal isolates during wastewater treatment using constructed trickling filters with charcoal and corn husks in D-Line, Port Harcourt, Rivers State, Nigeria. Microbiological analysis was done using Sabouraud Dextrose Agar (SDA) and Bushnell Hass Agar (BHS). The physicochemical results of both the contaminated water and the treated water using charcoal and corn husk show a major decrease in drinking water parameters like the Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Dissolved Oxygen (DO). The results obtained were 17.58 mg/l, 6.3 mg/l, and 5.25 mg/l; 31.30 mg/l; 10.9 mg/l, and 9.24 mg/l, 4.4 mg/l; 6.9 mg/l, and 7.0 mg/l, 6.84 mg/l; 6.64 mg/l, and 4.06 mg/l for contaminated water, charcoal treatment, and corn husk respectively. Fungal isolates found in the wastewater treatment include *Aspergillus niger*, *Aspergillus flavus*, *Yeast sp*, *Mucor sp*, *Rizopus sp*, *Geotrichum candidum*, *Aspergillus terreus*, and *saccharomyces cerevisiae* using corn husk filter. *Aspergillus niger* was the most prevalent species before treatment. The charcoal filter showed no fungal growth after 7 days. The constructed trickling filter using charcoal and corn husk were able to remove the color, reduce microorganisms, BOD, COD, offensive odour and some chemicals from wastewater. This constructed filter will help to treat wastewater fit for the purpose of irrigation and washing drilling equipment, thereby providing more of this scarce resource in areas it is needed. This answers to the United Nations Sustainable Development Goal (SDGs) 6.

Keywords: Wastewater, Reuse, Fungi, Filter, Physicochemical

INTRODUCTION

In developing nations like Nigeria, growing industrialization and urbanization have increased wastewater production, necessitating the urgent need for efficient treatment methods. Because of the dense population and lack of wastewater treatment facilities, managing wastewater is especially difficult (Odonkor & Ampofo, 2020). Organic debris, heavy metals, pathogens, and nutrients are among the many contaminants found in wastewater, which is generally produced by homes, businesses, and agricultural operations. The untreated or inadequately treated wastewater discharged into water bodies poses severe risks to human health and the environment, as it often contains harmful pathogens, including fungi, bacteria, and viruses (World Health Organization [WHO], 2020). Fungi are known to survive in extreme conditions, and some species can thrive in wastewater environments, leading to the contamination of treated effluents (Rahman et al., 2019). While fungi can play beneficial roles in wastewater treatment, some species are pathogenic and pose health risks. For instance, pathogenic fungi such as *Candida* and *Aspergillus* can thrive in wastewater environments, potentially causing infections and other health problems such as respiratory, skin, and systemic infections [4] (Iqbal et al., 2018). This effluent can contaminate water bodies if it is not properly handled, endangering the environment and public health (Rahman et al., 2019).

A rapidly expanding issue facing our world today is the constant inaccessibility of water, a vital resource for all life forms (Kummu et al., 2016). Some have linked the rising demand for freshwater for industrial, agricultural, and municipal purposes as well as climate change to the current water shortage (Ofori et al., 2021). Therefore, investigating the role of fungi in wastewater treatment systems is critical.

In recent years, concerns about increasing wastewater production have led to the development of innovative treatment methods, such as constructed trickling filters. These systems rely on biological processes to degrade pollutants and have been widely adopted due to their cost-effectiveness and operational simplicity. The trickling filter operates by allowing wastewater to flow over a bed of porous materials, on which microbial biofilms form and degrade organic contaminants (Ewing et al., 2018; Metcalf & Eddy, 2014). Wastewater treatment is essential for managing water pollution, protecting public health, and maintaining a sustainable environment. It involves removing contaminants such as organic matter, chemicals, and pathogens from used water before it is returned to natural water bodies or reused in industrial and agricultural processes. Traditional wastewater treatment systems, such as those relying on chemical and mechanical processes, are often expensive and unsustainable in resource-limited settings. Moreover, the improper design and management of wastewater treatment plants can result in inefficiencies, allowing harmful organisms to persist in treated effluents (Iqbal et al., 2018). Fungi in wastewater poses a unique challenge. One major concern is whether these alternative media can effectively control the growth of pathogenic fungi while promoting the proliferation of beneficial microorganisms.

It has previously been proposed to use treated wastewater effluent for irrigation as a substitute supply of water for farming (Ibekwe et al., 2018; Vergine et al., 2017a, Ofori et al., 2021, Jaramillo & Restrepo, 2017). This is because nearly any water quality can be produced, thanks to recent developments in wastewater treatment technologies. Determining which components need to be eliminated and to what degree remains the primary human and environmental problem (Shoushtarian, F., & Negahban-Azar, M. (2020).

The conventional wastewater treatment systems available in Rivers State are either insufficient or inefficient, often unable to handle the large volumes of wastewater generated daily (Ogolo et al., 2020). These systems also struggle to remove complex pollutants, including heavy metals, organic pollutants, and pathogens such as fungi (Nnadi & Obinna, 2021).

Traditionally, materials like gravel, stones, or plastic media have been used in trickling filters to support biofilm growth. Microbial processes degrade pollutants. As wastewater is passed over the filter media, biofilms of bacteria, fungi, and other microorganisms develop on the surface of the medium, facilitating the breakdown of organic materials. While traditional filter media like gravel and plastic have been effective, there is growing interest in finding sustainable and low-cost alternatives, especially in resource-limited situations. The use of charcoal and corn husks as filtration media has gained attention due to their potential advantages over conventional materials (Oyeleke et al., 2019).

Charcoal, which is derived from organic materials such as wood, has a porous structure that offers a large surface area for microbial colonization. This enhances the degradation of organic pollutants and the adsorption of heavy metals, making charcoal an effective filter medium (Dada et al., 2015). Additionally, charcoal is abundant and inexpensive, especially in regions like Rivers State, where it is produced as a byproduct of agricultural and forestry activities. Several studies have demonstrated charcoal's ability to improve water quality by reducing contaminants such as lead, mercury, and organic pollutants (Adie & Osibanjo, 2009). Similarly, corn husks, a byproduct of maize production, have shown promise as a low-cost and biodegradable filtration medium. Corn husks are rich in cellulose, hemicellulose, and lignin, which can adsorb organic and inorganic pollutants from wastewater. Their fibrous structure allows for effective filtration, while their natural composition supports microbial activity, including the growth of fungi and bacteria that degrade organic matter (Mohammed *et al.*, 2020). The utilization of corn husks in wastewater treatment systems is not only cost-effective but also contributes to waste management by repurposing agricultural byproducts (Oyeleke et al., 2019).

Named for the many rivers that cut across its terrain, Rivers State is a state in southern Nigeria. Large bodies of water including the Bonny, New Calabar, and Sombreiro rivers, as well as creeks, estuaries, and wetlands, may be found in this coastal state. The state's economy depends heavily on these water systems, which offer opportunities for transportation, fishing, agriculture, and oil exploration (Amadi et al., 2019). The state still has a lot of water resources, but access to clean water and water pollution are major problems that are made worse by inadequate wastewater treatment methods. If this issue is not resolved, fungal infection outbreaks and a decline in public and environmental health may result. It is critical to evaluate the fungal isolates in wastewater treatment systems that use charcoal and corn husks as filter media.

There is a growing interest in identifying locally available and affordable filtration materials that can perform as well as, or even better than, these expensive alternatives. Constructed trickling filters promote the growth of microbial biofilms that can break down organic matter and remove harmful pathogens from wastewater. These systems are relatively simple to operate, cost-effective, and can be adapted to local conditions, making them an ideal choice for regions like Rivers State, where infrastructure development may be limited (Ogolo et al., 2020). The use of charcoal and corn husks as filtration media is a promising solution due to their availability, low cost, and environmental sustainability (Nnadi et al., 2021). According to (Ajao & Anetor (2020), agriculture is a key source of employment in the state, with a large percentage of the population engaged in activities like irrigation farming, which relies on river water to irrigate crops. Similarly, fishing is a crucial part of the economy, and many communities depend on rivers and creeks as their primary source of fish and seafood (Amadi et al., 2019). In the absence of reliable municipal water supply systems, these natural water sources have become indispensable to local communities (Ilechukwuet al., 2021). The Bonny River, for instance, provides essential services to the Bonny Island population and other areas in Port Harcourt.

This work aims to study the fungal isolates and physicochemical characteristics of filtrates isolated from wastewater using charcoal and corn husks as filter materials.

2. MATERIALS AND METHODS

2.1 Study Area

The sampling site is located within the areas of D-Line in Port Harcourt. The geographical coordinates of the sampling location are 4.7783° N, 7.0085° E. The site is characterized by open drains that carry untreated wastewater from nearby markets, restaurants, and residential areas. The sources of waste are fruits, vegetables, and cleaning agents such as detergents. The wastewater from this location flows into local water bodies, making it a significant source of environmental pollution.

2.2 Materials Used and Their Treatment

The primary filtration materials used in the study were charcoal and corn husks. Both materials were selected based on their adsorption and filtration properties.

1. Charcoal: It was purchased from a local supplier in Mile 3 market, Port Harcourt, Nigeria. The charcoal was activated in the laboratory by washing, steeping in water, air drying and sterilize at 800°C for 1 hour in a muffle furnace to increase its porosity and adsorption capacity.
2. Corn Husks: The corn husks were sourced from a local farm in Port Harcourt, Nigeria. The corn husks were washed, steeped in 95 % alcohol, sun-dried, before being used as a filtration medium in the trickling filter.

3. Pebbles: Pebbles were bought from Mile 3 market, it was washed and sterilized before putting it inside the setup
4. Gravel: Gravels were bought from building sites. They were properly washed, air dried and sterilized microbes before putting them inside the setup.
5. Sharp sand: sharp sand was also gotten from building site and washed, air dried, and sterilized before putting it inside the setup

The set up was arranged in a vertical order in a recycled dispenser bottle. A tap was constructed at the base of the bottle to collect filtrates, and the opening was covered with a cork made from cotton wool and foil paper. The bottle was wrapped with black duct tape to avoid direct sun light and placed on a tabletop in the laboratory.

2.3 Water Sample Collection

Water samples were collected from the open drains in the sample area using 20-liter cans. The sampling was done at three different points along the drain to ensure a representative sampling. The water samples were placed on ice packs and immediately transported to the laboratory to preserve their integrity for microbiological and chemical analysis.

2.4 Physicochemical Analysis

The analysis done for the wastewater before and after were Dissolved oxygen (DO), biochemical oxygen demand (BOD), chemical oxygen demand (COD), turbidity, pH, temperature, Total Dissolved Solids (TDS), Total Suspended Solids (TSS), THC

2.5 Microbiological Analysis

2.5.1 Total Heterotrophic Fungi (THF)

The total heterotrophic counts (THC) of fungi (THF) in the wastewater were determined using the spread plate method. Serial dilutions of the water samples were prepared, and aliquots of 0.1 ml were placed onto the media (Sabouraud dextrose agar, SDA) in replicates using spread plate technique. The plates were incubated at 28°C for 5-7 days. Colony-forming units (CFU) were counted, and the results were recorded as CFU/mL

2.5.2 Hydrocarbon Utilizing Fungi (HUF)

The Bushnell-Haas Mineral Salt Medium is specifically formulated for the enumeration, isolation, and identification of hydrocarbon-utilizing microbes. It was supplemented with Streptomycin to support only the growth of fungi degrading hydrocarbon. The vapor phase technique was employed in isolating fungi from samples in replicates, it was incubated at room temperature for 3-7 days.

2.6 Morphological Analysis

Fungal isolates were identified by observing spore structures and colony characteristics such as color, texture, and size.

2.7 Microscopy

Fungal microscopy was performed using lactophenol cotton blue staining to observe hyphal structures and spore morphology under the microscope.

3. RESULTS

Table 1: Physiochemical Characterization of the contaminated water sample

Parameter	Unit	Contaminated Water Replicates			Average
General Appearance	-	Not Clear	Not Clear	Not Clear	
Color	-	Slightly Cloudy	Slightly Cloudy	Slightly Cloudy	
Odor	-	Objectionable	Objectionable	Objectionable	
pH	-	6.84	6.85	6.85	6.85
Temperature	°C	28.5-	28.5	28.5	28.5
TDS	mg/L	149	149	149	149
TSS	mg/L	0.082	0.084	0.085	0.83
DO	mg/L	4.3	4.4	4.4	4.4
BOD	mg/L	17.95	17.70	17.71	17.79
COD	mg/L	31.592	31.152	31.170	0.436
THC	mg/L	126	126	124	126

Key: DO (Dissolve Oxygen), BOD (Biochemical Oxygen Demand), COD (Chemical Oxygen Demand), THC (Total Hydrocarbon Content), TS(Total Solids), Total Dissolved Solids (TDS).

Table 2: Physiochemical Characterization of the corn husk sample

Parameter	Unit	Contaminated Water Replicates			Average
General Appearance	-	Not Clear	Not Clear	Not Clear	
Color	-	Slightly Cloudy	Slightly Cloudy	Slightly Cloudy	
Odor	-	Objectionable	Objectionable	Objectionable	
pH	-	4.06	4.04	4.06	4.05
Temperature	°C	31.0	31.0	31.0	31
TDS	mg/L	495	495	495	495
TSS	mg/L	0.180	0.180	0.180	0.180
DO	mg/L	7.0	7.0	7.0	7.0
BOD	mg/L	5.50	5.25	5.00	5.25
COD	mg/L	9.68	9.24	8.80	9.24
THC	mg/L	30.56	30.56	30.56	30.56

Key: DO (Dissolve Oxygen), BOD (Biochemical Oxygen Demand), COD (Chemical Oxygen Demand), THC (Total Hydrocarbon Content), TSS (Total Suspended Solids), Total Dissolved Solids (TDS)

Table 3: Physiochemical Characterization of the charcoal filtered water

Parameter	Unit	Contaminated Water Replicates			Average
General Appearance	-	Clear	Clear	Clear	
Color	-	Colourless	Colourless	Colourless	
Odor	-	Unobjectionable	Unobjectionable	Unobjectionable	
pH	-	6.64	6.64	6.64	6.64
Temperature	°C	31.0	31.0	31.0	31
TDS	mg/L	443.08	443.08	443.08	443.08
TSS	mg/L	0.084	0.084	0.084	0.084
DO	mg/L	6.9	6.9	6.9	6.9
BOD	mg/L	6.25	6.00	6.75	6.33
COD	mg/L	11.00	10.56	11.00	10.85
THC	mg/L	26.99	26.99	27.06	27.01

Key: DO (Dissolve Oxygen), BOD (Biochemical Oxygen Demand), COD (Chemical Oxygen Demand), THC (Total Hydrocarbon Content), TSS (Total Suspended Solids), Total Dissolved Solids (TDS)

Table 4: Fungal Isolates of Contaminated Water

Analysis	Replicates		Average
THFC	20×10^4	16×10^4	1.8×10^4
HUF	4.0×10^4	6.0×10^4	5.0×10^4

Table 5: Fungal Isolates of Corn Husk filtrates

Analysis	Replicates		Average
THFC	6.0×10^4	8.0×10^4	7.0×10^4
HUF	3.0×10^4	4.0×10^4	3.5×10^4

Table 6: Fungal Isolates of Charcoal filtrate

Analysis	Replicates		Average
THFC	-	-	-

HUF

Table: 7: Percentage Occurrence of Fungal Isolates from Contaminated Wastewater

Isolates occurrence	Frequency of occurrence	Percentage of occurrence (%)
<i>Aspergillus flavus</i>	7	12.5
<i>Rizopus sp</i>	5	8.9
<i>Mucor sp</i>	7	12.5
<i>Saccharomyces cerevisiae</i>	5	8.92
<i>Aspergillus niger</i>	10	17.86
<i>Yeast sp</i>	6	10.7
<i>Geotrichum candidum</i>	8	14.3
<i>Aspergillus terreus</i>	8	14.3
TOTAL	56	100

Table8: Percentage Occurrence of Fungal Isolates from Corn Husk Filter

Isolates occurrence	Frequency of occurrence	Percentage of occurrence (%)
<i>Aspergillus flavus</i>	7	38.89
<i>Rizopus sp</i>	3	16.67
<i>Mucor sp</i>	5	27.78
<i>Saccharomyces cerevisiae</i>	3	16.67
TOTAL	18	100

Table9: Percentage of Occurrence of Fungi Isolates before and after treatment

Isolates	(Before)	(After)
<i>Aspergillus flavus</i>	12.07	15.15
<i>Aspergillus niger</i>	21.21	21.21
<i>Geotrichum candidum</i>	13.79	5.17
<i>Rizopus sp</i>	12.07	12.12
<i>Aspergillus terreus</i>	13.79	15.15
<i>Mucor sp</i>	12.07	5.17
<i>Yeast so.</i>	10.34	5.17
<i>Saccharomyces cerevisiae</i>	8.62	5.17

3.1 Physicochemical characterisation of samples

The general appearance of the polluted water sample was not clear, the colour was cloudy, the odor was objectionable, the pH was 6.85, temperature was 28.5 degrees. The chemical analysis was 149 mg/l, 0.83 mg/l, 4.4 mg/l, 17.79 mg/l, 0.436 mg/l, and 126 mg/l for TDS, TSS, DO, BOD, COD, and THC respectively as shown in Table 1.

The general appearance of the corn husk filtered water sample was not clear, the colour was cloudy, the odor was objectionable, the pH was 4.05, temperature was 31 degrees. The chemical analysis was 495 mg/l, 0.180 mg/l, 7.0 mg/l, 5.25 mg/l, 9.24 mg/l, and 30.56 mg/l for TDS, TSS, DO, BOD, COD, and THC respectively as shown in Table 2.

The general appearance of the charcoal filtered water sample was not clear, the colour was cloudy, the odor was objectionable, the pH was 6.64, temperature was 31 degrees. The chemical analysis was 443.05mg/l, 0.084mg/l, 6.9mg/l, 6.33 mg/l, 10.85 mg/l, and 27.01 mg/l for TDS, TSS, DO, BOD, COD, and THC respectively as shown in Table 2.

The pH range in the charcoal filter agrees with the EPA British Columbia range of 6.0 – 9.0 and so can be used for irrigation.

3.2 Characterization of fungi

The total heterotrophic fungi obtained for contaminated water was 1.8×10^4 while the hydrocarbon utilizing fungal count was 5.0×10^4 as seen in Table 4.

The total heterotrophic fungi obtained for corn husk filtered water was 7.0×10^4 while the hydrocarbon utilizing fungal count was 3.5×10^4 as seen in Table 5.

No fungal growth was observed in charcoal filtered water after 7 days (Table 6).

The fungal isolates obtained from the samples was *Aspergillus niger*, *Mucor racemosus*, *Fusarium spp*, *Saccharomyces*, *Aspergillus fumigatus*, *Yeast spp*, *Aspergillus flavus*.

The percentage occurrence of organisms in contaminated water was *Aspergillus flavus* 12.5 %, *Rizopus* sp 8.9 %, *Mucor sp* 12.5 %, *Saccharomyces cerevisiae* 8.92%, *Aspergillus niger* 17.86 %, *Yeast sp* 10.7 %, *Geotrichum candidum* 14.3%, *Aspergillus terreus* 14.3 % as seen in Table 6.

The percentage occurrence of organisms in corn husk filtered water was *Aspergillus flavus* 38.89 %, *Rizopus* sp 16.67 %, *Mucor sp* 27.78 %, *Saccharomyces cerevisiae* 16.67 % (Table 8).

3.3 Discussion

The pH range in the charcoal filter agrees with the Environmental Protection Agency of British Columbia range of 6.0 – 9.0 and so can be used for irrigation. The pH range for corn husk treated water is slightly acidic which suggests that it is not fit for irrigation as it may result in nutrient imbalance which may alter plant growth (de Souza Santos et al, 2011). It also suggests that it will not be fit for washing drilling equipment and cooling of drilling bits as it may lead to corrosion of the pipes and sprinklers (Jeong and Jang, 2016). The result of the charcoal filtered water was clear, colourless, and unobjectionable. This meets the standard for recycled water for irrigation and other purposes (Metcalf et al, 2007; Shoushtarian & Negahban-Azar, 2020).

The polluted water wasn't clear, and it had an offensive smell which can be attributed to the decomposing organic waste in the water and inorganic chemicals in the water (Metcalf et al, 2007). The total dissolved solids were 149 mg/l, 495 mg/l, and 443.05 mg/l for contaminated water, corn husk filtered water and charcoal filtered water. This increase is because of the addition of particles the filter material contributed to the dissolved solids. This trend was observed in the TSS, dissolved solids, and COD while a reduction trend was observed in the BOD and THC (WHO, 2006). This agrees with (Shoushtarian & Negahban-Azar, 2020).

The water treated using only one step filtration with organic materials corn husk and charcoal is very efficient in removing fungal load, odour, colour, and chemical composition of contaminated water. This means that this setup can be used in water recovery and reuse in areas with water supply shortage. These filtrates can be used in irrigation ((Shoushtarian &Negahban-Azar, 2020), toilet channels, recreational purposes, and even domestic use (Shoushtarian, &Negahban-Azar, 2020).

Water quality of recycled water is usually categorised into three main groups: human health parameters, agronomic parameters (toxic ions, trace elements, nutrients, and salinity), and physicochemical parameters such as turbidity, TSS, BOD, and COD. The filtrate from this charcoal filtration meets the physicochemical, and human health parameter standard for irrigation). The categories of human health and physicochemical parameters were analysed in this study, and they met standards for irrigation (Shoushtarian &Negahban-Azar, 2020).

This will reduce the lack of fresh water due to population growth in areas with poor water supply (Jeong & Jang, 2016).

CONCLUSION

With the recent advances in technology, wastewater can be treated to almost any quality. Although there are a few concerns about how much microbes, chemicals, and physical constituents these treatments can remove, the major concerns are the safety of water reuse. This study showed that toxic substances and fungal loads can be significantly reduced in water using charcoal and corn husk in just a one-step constructed filtration setup.

The microbial, physical, and chemical quality of water varies according to the purpose for its use (Lazarova& Bahri,2004, Poonam et al, 2013). The filtrate from this charcoal filtration meets the physicochemical, and human health parameter standard for irrigation ((Shoushtarian &Negahban-Azar, 2020). Further treatment can get the filtrate to drinking water quality (Shoushtarian, F., &Negahban-Azar, M. 2020).

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