TREATMENT OF SULLAGE USING ACTIVATED CARBON FROM CONSORTIUM OF SELECTED AGRO-WASTES

BY

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CERTIFICATION

s is to certify that this work: "Treatment of Sullage Using Activated Carbon From nsortium of Agrowastes" was carried out by CHIDIMMA ADAMMA NDUKA 164993438) in partial fulfilment for the award of the degree of M.Sc. in Biotechnology in the partment of Biotechnology of the Federal University of Technology, Owerri.

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DEDICATION

This work is dedicated to my father Late Engr. G. I. Nduka, and mother Mrs. S. A. Nduka, my aunt, Mrs. J. Nduka and my siblings for their contributions in my life.

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ABSTRACT

The study investigated the effectiveness of treating sullage using activated carbons from rice husk, corn cob and coconut husk. The agrowastes were sourced from Relief Market, Owerri while sullage samples were collected with 10 litre container from Federal University of Technology, Owerri (FUTO) students' hostels. Agrowastes were carbonized at 600^oC and chemically activated using phosphoric acid (H_3PO_4) before they were used as adsorbents. pH, Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), nitrate (NO_3^-), phosphate (PO_4^-), chloride (Cl^-), sulphate (SO_4^{2-}); heavy metals (mercury, Hg; cadmium, Cd; chromium, Cr and lead, Pb)) and microbial load of the sullage samples were analyzed before and after treatment with biosorbents. Titrimetric, spectrophotometric, and cultural methods were used to determine the chemical, heavy metals concentrations and microbial load respectively. Percentage yield of activated carbon were 35.7% (rice), 22.0% (corncob) and 10% (coconut husk). Initial concentration of the chemical parameters of sullage samples showed COD to be 486.2±87 mg/l, BOD, 175.0±3.1 mg/l; pH, 7.33±0.1; Cl^-, 31.0±5.5 mg/l; NO_3^-, 28.6±24 ppm; PO_4^-, 7.08±3.2 ppm and SO_4^{2-}, 347.08±67.1 ppm. After treatments, COD ranged from 122.2±10 - 190.5±25 mg/l, BOD, 44.0±3.5 – 68.6.0±9.0 mg/l, pH, 7.08±0.1 - 7.18±0.1, Cl^-, 17.28 - 21.13 mg/l, NO_3^-, 0.0 - 12.79 ppm and sulphate 117.7 - 251.7 ppm. Initial values of heavy metals were: Hg 4.01±0.6 ppm, Cr, 0.13±0.3, Cd, 0.076±0.0 ppm and Pb, 0.06±0.0 ppm. After treatments, Hg ranged from 1.60±0.5 - 2.51±0.3, Cr, 0.00; Cd, 0.0 – 0.008 ppm, Pb, 0.0 – 0.06 ppm. Before treatment, Total Heterotrophic Count (THC) was $1.2x10^{11}$ cfu/ml, Total Coliform Count (TCC), $6.4x10^6$ cfu/ml and Total Fungi Count (TFC) $2.2x10^{10}$ cfu/ml. *Staphylococcus* sp, *Micrococcus* sp, *Bacillus* sp, *Salmonella* sp, *Saccharomyces* sp and *Penicillium* were present in the sullage. THC after the treatment ranged from $1.69x10^9$ to $7.6x10^{10}$ cfu/ml; TCC, $2.2x10^5$ - $7.3x10^8$ cfu/ml and TFC $1.0x10^8$ - $1.2x10^9$ cfu/ml. There was significant difference ($p \leq 0.05$) between the untreated and the treated sullage samples. The efficiency of the individual adsorbents in reducing the chemical parameters was of this order: ricehusk > corncob > coconut husk. In combination it was, rice husk+corncob+coconuthusk > ricehusk +corncob > corncob+coconuthusk > ricehusk+coconuthusk. For heavy metals removal, efficiency of individual adsorbents was of the order: ricehusk > corncob > coconuthusk, while when combined, it was: ricehusk+corncob > corncob+coconuthusk > ricehusk+corncob+coconuthusk > rice husk+coconut husk. High percentage reduction observed of chemical properties, heavy metal levels; complete elimination of *Bacillus* sp and general reduction of the microbial load revealed that activated carbons from rice husk, corn cob and coconut husk can be used singly or combined for the purification of sullage.

Keywords: Activated Carbon, Agrowastes, Carbonization, Coconut Husk, Corn Cob, Rice Husk, Sullage.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND INFORMATION

One of most valued resource for the existence of all living organisms is water. However, this valued resource is increasingly being threatened as human populations grow and demand more water top quality for domestic functions and economic activities (David & Omoogun, 2016). Due to man's anthropogenic activities a lot of substances have been built up in water to cause physical, chemical and/or biological change to an extent that they cause problems for animal, man and the environment. These substances are often in form of contaminants or pollutants. When such pollutants are introduced into any water body, they cause a situation regarded as water contamination or pollution (Owa, 2013). Be it surface water pollution (pollution of rivers, lakes and oceans) or ground water pollution, all water pollution is dangerous to the health of living organisms. Pollutants may be organic or inorganic and maybe in solid or liquid forms (wastewater). Organic pollutants include detergents, disinfection by-products (chloroforms), insecticides, herbicides, petroleum, volatile organic compounds, chlorinated solvents, chemical compounds found in personal hygiene, cosmetic compounds and drug compounds. Inorganic water pollutants include acidity caused by industrial or domestic discharge (sulfur dioxide from power plants, ammonia from food processing waste, and chemical waste as industrial by-products, fertilizers, heavy metals etc).

Water pollution is a major world drawback and it needs ongoing evaluation and revision of water resource policy at all levels. At global level, around 80% of wastewater is discharged into the environment untreated causing widespread water pollution (United Nations World Water Assessment Program (UN WWAP, (2009); Rosenthal (2005). These result in the contamination of water supplies and ground waters which are used for numerous activities like irrigation, drinking and recreational activities. According to USGS (2017) there is hardly any non-communicable public health outbreak without a link to environmental contamination, arising from daily human interaction with the environment. Like other developing countries, the management of wastewater has been one of the major problems in Nigeria. Major problems leading to wastewater pollution in Nigeria result from population, industrialization and urbanization. Major sources of wastewater are from industries and domestic activities and have entailed a tremendous increase in discharge of a wide diversity of pollutants to receiving water bodies.

Domestic wastewater categorized as organic pollutant includes grey water or sullage (wash water from dishwashers, washing machines sinks, bath tubs etc.) and black water (fecal sewage); they accounts for 67.5% and 32.5% of domestic waste water respectively. Domestic wastewater when not properly managed can cause undesirable effects on the different components of the aquatic environment and on fisheries. Fresh water pollution can be detrimental to the health of humans and aquatic organisms as it is used as primary sources of water by population all over the world particularly in Nigerian communities (Abdulmumini, Gumel & Jamil, 2014).

In Owa's (2013) report, water contamination accounts for nearly 14,000 mortality cases reported daily in developing countries. In October 2010, 29,115 cases involving 1,191 deaths of epidemic cholera have been reported in just 15 out the 36 States including Federal Capital Territory. The figure increased respectively from 1616 and 126 deaths in 2004. Consistent with Galadima *et al.* (2011) it had been discovered that the happening remains existing in new areas due to continuous water pollution. A study by Umeh *et al*. (2004) showed that 48% of the people in Katsina-Ala Local Government area of Benue state are affected by urinary schistosomiasis, due to increase in water pollution index. Studies done by Alens (2014) and Environmental Protection Agency, EPA (2015), revealed that sewage pollution alone contributes to significant cases of aquatic live poisoning, inflammatory disease, hepatitis and infections in the eyes or nose during dermal exposure. Studies (Ibeto & Okoye, 2010) conducted on 240 people, comprising of children, pregnant/nursing women and men in Enugu State showed some heavy metal poisoning that resulted from absorption of contaminated water or via associated food. Nickel, chromium and manganese were detected with concentrations exceptional the allowed limits permissible by WHO, within the blood samples of the respondents. The poisoning was believed to be occupational and water-based. In a related development, more than 400 children from seven villages around Bukkuyum and Gummi Local Government areas of Zamfara state, died from lead poisoning within six months in 2010. The poisoning primarily associated with mineral exploitation, consumption of water and food and air-based inhalation, have so far affected 3,600 children, with further expectations that 180 villages covering around 30,000 people may be affected.

In most countries, the principal risks to human health related to the consumption of polluted water are microbiological in nature (WHO, 2011). The most important waterborne diseases are typhoid, cholera, bacterial and amoebiasis, enteritis, polio**,** hepatitis A, and schistosomiasis. Other health problems are poor blood circulation, skin lesions, vomiting and damage to the central nervous system (Asubiojo, 2016). The bacteriologic examination of water features a special significance in pollution studies, as it is a direct measurement of deleterious effect of pollution on human health (APHA, 2005). Coliforms are one of the microbial indicators for monitoring water quality, other microorganisms generally found in surface waters that have caused human health issues include: *Staphylococcus* spp, *Shigella* spp, *Cryptosporidium parvum*, *Giardia lamblia*, *Salmonella, Novovirus* and other viruses, parasitic worms (Brenner *et al.*, 1993; Grant, 1997). Collection, treatment and disposal of industrial and domestic wastewater are the serious issues to be handled (Ezeronye & Amogu, 1998).

According to Kolawole *et al.* (2011) wastewater management and prevention of water pollution requires effective monitoring of physico-chemical and microbiological parameters, proper education and enlightenment of local people on the importance of water sanitation and good waste disposal method. Strategies for wastewater management for centuries had relied on the self purification mechanisms (Singh, Abdullah & chhotu, 2018). Land, water and air were assumed to absorb unlimited amounts of wastes without adverse impact on living things. Though this self purification mechanism has provided some relief at the past, the present discharge and concentrations exceed effective thresholds of the natural ecosystem (Nuhu, Omali & Clifford, 2018). Coagulation, filtration with coagulation, precipitation, ozonation, absorption, ion exchange, reverse osmosis, advanced oxidation and alternative new strategies were later employed however they were found to be limiting since they involve high capital, operational costs, space wastage, and they are commercially not pleasing in nature, with much of disposal issues. Among the potential techniques employed, adsorption was found to be less expensive and ecofriendly.

Activated carbon also known as activated coal, a carbon based compound that have been processed to make it extremely porous have varied applications in removing pollutants from air or water such as in groundwater remediation, drinking water filtration, air purification and volatile organic compounds capture from painting and other processes (Nuhu, Omali & Clifford, 2018). Its usefulness derives from its large micropore (and sometimes mesopore), volume and high surface area. The quest for economically safe activated carbon to be used as adsorbents has lead to the exploration of biomass (rice husk, saw dust, oil palm shell etc) for the removal of pollutants from wastewater (Nasin, Shaliza & Piarapakaran, 2004). Agrowastes have great potentials as inexpensive adsorbents (Nasin, Shaliza & Piarapakaran, 2004). Their abundance and handiness make them good sources of materials for activated carbon (Rahmani, Mahvi, Vaezi, Mesdaghinia, Nabizade, & Nazmara, 2009). Many carbonaceous materials such as bark, shells, husk, etc are used in the production of commercial activated carbon (Tadda *et al*., 2014). This study will determine some physicochemical properties, heavy metals and other contaminants found in the male and female section of the FUTO Student hostels and explore on the potential of activated forms of rice husk coconut husk and corn cob as adsorbents to treat the sullage.

1.2 PROBLEM STATEMENT

Most of the freshwater bodies in Nigeria are daily taking in varieties of pollutants from household sullage (Pachkor & Parbat, 2017). The daily amount generated is 36,493, 920 liters (36494m^3) (Abdulahi, Humuani & Musa, 2013). All sources and forms of water pollution are dangerous to the health of living organisms. Much attention has been given to industrial wastewater pollution while the effect of domestic/ household wastewater is undermined. Domestic water is the primary source of pathogens and organic substances because most pathogens are excreted in feces and discharged. It is estimated that up to 80 percent of all domestic wastewater is sullage, and they are from individual houses, cities and towns and are likely to contain pathogens of some type, potentially presenting a direct threat to public health. Domestic wastewater when not properly managed can cause undesirable effects on the different components of the aquatic environment, on fisheries, man and other life supporting systems.

Adsorption is recognized as a good and low value technique for the removal of organic pollutants from water and wastewater, and produce high-quality treated effluent. Activated carbons are common adsorbents used for the removal of undesirable odor, taste, color, and other inorganic and organic impurities from domestic and industrial wastewater attributable to their massive extent, small porous structure, non polar character and because of its economic viability (Tadda *et al*., 2014). But it may be hard to establish individual adsorbent that are more suitable for the treatment of household wastewater (sullage). This engendered the need to carry studies on treatment of sullage using activated carbon from of rice husk, corn cob and coconut husk.

1.3 AIM AND OBJECTIVES

This research aimed at treating sullage with activated carbon from selected agrowastes (rice husk, corn cob and coconut husk).

1.3.1 OBJECTIVES

- To determine the percentage yield of activated carbon.
- To determine the chemical properties (Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), pH, chloride, nitrate, sulphate and phosphate of the sullage before and after treatment with activated carbon from agrowastes.
- To determine the heavy metal concentration (chromium, lead, cadmium and mercury) of the sullage before and after treatment with activated carbon from agrowastes.
- To determine the microbial properties of the sullage before and after treatment with activated carbon from agrowastes.
- To compare the efficiency of the activated forms of the adsorbents in treating sullage.

1.4 HYPOTHESES

This study was guided by the following hypotheses:

1. There is no difference in the chemicals, heavy metals and microbial properties of the sullage from male, female and their combination from FUTO hostel.
2. Activated carbon from rice husk, corn cob and coconut husk does not reduce the chemical, heavy metals, microbial properties of sullage, in singly and combined forms.

1.5 JUSTIFICATION OF STUDY

The study assessed the novel ways of using indigenous agro-wastes in their activated forms for the treatment of sullage and it is hoped that the results of this study will assist individuals and tertiary institutions in using these adsorbents in the treatment of sullage effluent before discharge into the environment.

1.6 SCOPE OF STUDY

This research work determined the efficiency of individual and consortium of activated carbon from rice husk, corn cob and coconut husk in the treatment of sullage through assessment of the sullage's chemical, inorganic compounds and microbial properties before and after treatment.

CHAPTER TWO

LITERATURE REVIEW

2.1 CLASSIFICATION OF WASTEWATER

Wastewater is any water that has been negatively affected in quality by man is termed wastewater. It comprises liquid wastes that are discharged from domestic residence, commercial properties, industrial plants and agricultural facilities or lands (Abdalrahman & Alsulaili, 2015). Wastewater discharged from domestic residence is called domestic wastewater i.e. wastewater from the kitchen, shower, wash basin, toilet and laundry (Vinnerås, Palmquist, Balmer, & Jonsson, 2006). Domestic wastewater is transported through a physical infrastructure known as sewerage which leads to septic tanks or gutters (channeled to streams and rivers).

According to Samwel (2005), domestic wastewater is classified as black water and grey water/sullage. Blackwater is used to describe wastewater from toilets which likely contain pathogens. Wastewater from domestic equipment other than toilet is called greywater or sullage. Sullage includes wastewater from the sinks, showers, baths, and laundry machines or dish washers. Sullage could also be contaminated with excretory product through washing of person's anal or from washing underwear and nappies and dead skin cells, therefore are not free from pathogens. In some sanitation systems, it is preferred to keep the sullage separate from blackwater to reduce pollution and to simplify treatment methods for the sullage (Tilley *et al*., 2017).

2.2 COMPOSITION AND CHARACTERIZATION OF SULLAGE

The composition and characterization of wastewater are classified into physical, chemical and biological.

Physical / general quality

The amount of wastewater generated within a house varies greatly and depend on several factors such as the age and number of occupants, their habits and how they use water. Some European cities can reach to up 586 L/ day wastewater generated. According to Pachkor & Parbat (2017) sullage accounts for 68% of the total wastewater generated, mainly composed of baths and showers and laundry. Physically, sullage is usually characterized by a grey colour, musty odor and has a solid content of about 0.1%. The solids can be suspended as well as dissolved. Sullage contributes significantly to wastewater parameters such as biochemical oxygen demand (BOD), chemical oxygen demand (COD), total suspended solids (TSS), ammonium (NH_4^+), total phosphorous, boron, metals, salts, surfactants, synthetic chemicals, oils and greases, xenobiotic substances, microorganisms etc. (Wiel-Shafran *et al.,* 2006; Travis *et al*., 2008; Gross, Kaplan & Baker, 2007; Eriksson & Donner, 2009). Untreated domestic wastewater typically contains 50 to 100 mg/L of oils and greases with approximately 2/3 of the load contributed by sullage (Gray & Becker, 2002; Tchobanoglous *et al*., 2003). All of these components have potential negative environmental and health impacts.

Chemical quality

Chemical contamination found in bathroom sullage originates from shampoo, hair dyes, toothpastes and cleaning chemicals. Laundry water contains higher chemical concentrations from soap powders and soiled clothes (sodium, phosphate, boron, ammonia, nitrogen), and is high in suspended solids, lint, turbidity and oxygen demand and if applied to untreated land could lead to environmental damage, as well as threat to public health (WHO, 2006). Chemically, sullage is composed of organic and inorganic compounds as well as various gases, like other type of wastewater (Pachkor & Parbat, 2017). Organic compounds consist primarily of carbohydrates, proteins and fats which reflect the diet of the people. Inorganic components include sulphur, nitrogen, phosphorus, pH, chlorides, alkalinity, heavy metals, and toxic compounds etc. Other chemical properties of sullage include chemical Oxygen Demand, Biochemical Oxygen Demand (Erikson & Donner, 2009). Sullage contains significant amounts of nutrients, particularly nitrogen and phosphorus. An average volume of sullage (356 L per day) will produce approximately 45 g of nitrogen and 3 g of phosphorus per day (WHO, 2006). Test to ascertain water qualities are presented in Table 2.1.

Route of entry

Chemicals can enter an organism by various routes such as body surface, ingestion, and inhalation. If sullage is released directly to soil, the soil flora and fauna are exposed to the components of the sullage.

Table 2.1: Some important water quality tests, their major significance and general means of measurement

Quality parameter	Significance	General methods of analysis; expression of result
Colour (apparent)	Suspended and dissolved solids	Colorimetry method: comparison with platinum cobalt standard: unit of colour being produced by 1 mg/l platinum in the form of chroplatinate ion.
Odour	Most organic and some inorganic chemicals.	Subjective perceived odour: threshold number
Turbidity	Estimate of suspended matter	Jackson candle turbidometry (Jackson units) or Nephelometry method (Formazin units)
Dissolved oxygen	Potential for oxidation of organic matter; life support	Titrimetry or electrochemical method percent saturation or mg/l 0_2
Carbon dioxide	Aerobic/anaerobic decomposition of organic matter; carbonate equilibrium	Nomographic, titrimetry mg/l of CO_2
Total suspended solids	Turbidity: treatment efficiency	Gravimetry; mg/l
Settleable solids	Turbidity; treatment	Gravimetry; mg/l
Total dissolved solids	Salinity: may affect ecosystems and domestic and agricultural usefulness	Gravimetry; mg/l

Table 2.1: Some important water quality tests, their major significance and general means of measurement (continued)

Quality parameter	Significance	General method of analysis; expression of results
Total solids	General polluting potential	Gravimetry; mg/l
Calcium	Hardness: Scale formation	Tritimetry and gravimetry mg/l of $CaCo_3$
Total Organic Carbon (TOC)	Extent of organic matter	Tritimetry and colorimetry; mg/l of carbon
Organic nitrogen	Extent of nitrogenous organic matter	Colorimetry of titrimetry mg/l of organic nitrogen
Ammonia nitrogen	Extent of nitrogenous organic matter (proteins) toxicity	Titrimetry: mg/l of ammonia nitrogen
Nitrate nitrogen	Extent of oxidation of NH_3 plant nutrient may serve as source of 0_2, toxic in excessive amounts (Methaeomglobinemia)	Colorimetry: mg/l of ammonia nitrogen
Phosphate	Plant nutrient	
Sulphate	Possible reduction to H_2S: corrosion of concrete, possible gastrointestinal irritation	Colorimetry: mg/l of phosphate as PM_4^{3-}
Cyanides	Toxic potential	Gravimetry or colorimetry: mg/l of sulphate as SO_4^{2-}
Phenols	Toxic potential odour; taste	Colorimetry; mg/l or titrimetry: mg/l or cyanide
Permanganate values	Oxidizable inorganic matter, also oxidised organic matter	Titrimetry, chromatography or dolorimetry: mg/l of phenol
Synthetic detergents	Foam: toxic potential: taste	Titrimetry: mg/l or oxygen consumed from a standard permanganate solution
		Colorimetry or titrimetry: mg/l of specific detergent

Table 2.1: Some important water quality tests, their major significance and general means of measurement (continued)

Quality parameter	Significance	General method of analysis; expression of results
Pesticides	Toxic potential	Gas chromatography method: mg/l of pesticide
Iron	Taste; discolouration: turbidity; growth of (iron) bacteria	Colorimetry or atomic absorption spectroscopy: mg/l of iron
Magnesium	Hardness: taste, possible gastrointestinal irritation, scale formation	Colorimetry or atomic absorption spectroscopy: mg/l or magnesium
Hardness	Soap consummation; scale formation	Titrimetry: mg/l of CaCo3
pH value	Intensity of acid or alkali present, strength of effluents affects many chemical and biological properties	pH meter
Chloride ion	Degree of pollution, sewage; degree of salt water intrusion; taste, corrosion in her water systems	Titrimetry or colorimetry: mg/l of Cl^-
Stability/saturation with respect to calcium carbonate	Ability to maintain oxidised condition: tendency to revert to anaerobic conditions with foul odours	Stability/saturation index
Biochemical Oxygen Demand (BOD)	Extent of biodegradable organic matter	Measurement of dissolved oxygen before and after incubation for 5 days at 20^0C: mg of oxygen consumed per litre
Chemical Oxygen Demand (COD)	Organic matter susceptible to oxidation by a strong chemical oxidant	Titrimetry: mg/l of oxygen consumed from standard dichromate solution

Source: WHO, 2006

Biological / Microbial quality

Sullage if it contains human faeces may contain pathogens in the form of bacteria, viruses, protozoa, and/ parasites. In fact, some activities such as washing faecal contaminated laundry, childcare and showering, add faecal contamination to sullage (Ottoson & Stenström, 2003). Occasionally, gastrointestinal bacteria such as *Salmonella* and *Campylobacter* can be introduced by food-handling in the kitchen (Friedler, 2004). Sullage may have an elevated load of easily degraded organic material, which may favour growth of enteric bacteria (faecal indicators) and such growth has been reported in wastewater systems (Marville *et al.*, 2001). Laundry sullage exhibit a high range of the values of suspended solids, salts, nutrients, organic matter and pathogens which arise from washing of clothes using detergents. Kitchen sullage is reported as the highest contributor of oils and greases in domestic greywater, but oils and greases are present in all sullage streams (Friedler, 2004).

Routes of entry

The environmental transmission of pathogens occurs through several different routes. These may be directly through contact with greywater; directly through contaminated drinking-water; directly through vegetables, shellfish or other food products exposed to contaminated water or soil; by accidental ingestion of contaminated water during recreational activities; by inhalation of aerosols or dust due to irrigation with greywater; vector-borne transmission where the vector or the intermediate host breeds in water; and by secondary transmission through contact with infected individuals. (Abdalrahman & Alsulaili, 2015).

The disease-causing organisms in greywater are mainly transferred through contaminated hands, contact with broken skin, contact with contaminated items such as toys, garden implements, grass or soil, transmission by pest vectors such as rats, mice, flies, cockroaches and transmission by family pets.

Parameter affecting the characteristics of sullage

The composition of sullage depends on several factors, including sources and installations from where the water is drawn:

- Quality and type of the water supply (groundwater well or piped water)
- Type of distribution net for drinking water
- Type of distribution net for sullage (because of leaching from piping, chemical and biological processes in the biofilm on the piping walls)
- Activities in the household (lifestyle, custom and use of chemical products)
- Installation from which sullage is drawn (kitchen sink, bathroom, hand basin or laundry wash)
- Type of source: industrial or household
- Geographical location
- Demographics and level of occupancy
- Quantity of water used in relation to the discharged amount of substances.

2.3.0 IMPACTS OF SULLAGE

Environmental impact

The major disadvantage associated with infiltration of sullage is that the risk of contamination of the soil and receiving waters because of the comparatively high content of various kinds of pollutants (chemical compounds and microorganisms). Bathroom sullage is usually composed of hair, soap, shampoo, conditioner, toothpaste, body fats, oils, cleanup product, hair dye, nutrients and microorganism, together with unclean coliforms, whereas laundry sullage usually contains lint, oils, greases, laundry detergents, chemicals, soap, nutrients, salts and microorganism together with coliforms (Ormiston Associates Ltd, 2008). Some laundry detergents are high in salt (typically as filler in powders) and can tend to accumulate within the soil profile and cause soil structure collapse. Liquid detergents tend to have much lower salt content. Greywater from laundry can affect the soil characters and plant growth by raising salinity (Anwar, 2011). Saeed et al. (2015) conducted a study to evaluate the impact of household sullage (soap water) on growth of *Sesbania grandiflora L* and *Sesbania grandiflora* showed that higher concentration of detergents revealed discovered cytotoxic effects resulting in poor growth and development of root nodules which can result to ionic toxicity or modification of hydraulic properties of the soil. High levels of sodium will cause discoloration and burning of leaves, and may contribute toward an alkaline soil condition. In addition, high sodium can be toxic to certain plants and can prevent calcium from reaching them. Detergent and laundry products also contain other chemicals that are harmful to plants, such as boron, chlorides and peroxides. Concentrations slightly higher than those considered useful will cause injury or death to plants. Nitrogen, a necessary nutrient for plant growth is extremely beneficial as a supplement to landscape plants. Phosphorus is a necessary nutrient for plant growth, and is very beneficial as a supplement to landscape plants. Potassium is beneficial, especially in soil with high alkalinity. Sodium can act as a poison to plants by reducing the plant's ability to take up water from the soil. Chlorine is undesirable for plants in large amounts. Bleaches and detergents carry large amounts of chlorine.

Department of Local Government, DLG 1998 (NSW Health, 2000) identifies how domestic wastewater may harm the environment and is summarized as follows:

- By overloading the soil with nutrients;

- By surpassing the hydraulic loading of the soil with water causing run off of polluted water to storm water drains, rivers, streams etc;
- By varying the soil salinity;
- By varying the soil permeability;
- By modifying the soil pH;
- By changing the soil electrical conductivity;
- By varying the soil sodicity;
- By varying the soil ion exchange capacity;
- By varying the soil phosphorus sorption capacity;
- By changing the soil dispersiveness; and
- By contaminating the soil with chemical impurities that have an effect on the properties of the soil to assimilate nutrients or water.

Human health

Improper management of household wastewater has great impacts on the quality of water. Household wastewater is directly discharged into rivers and streams through drainage lines, and results in the release of toxic substances, decreases levels of dissolved oxygen, increase nutrient loads and bioaccumulation in aquatic life (Environmental Canada, 1997). Household wastewater is the principal vector by which a large number of communicable diseases are transmitted and spread in urban areas (Sien, 2001). Sullage can introduce a wide range of pollutants and microbial contaminants to water sources. This includes ground water pollution of wells and boreholes. According to Debre Markos Town Health Centre Annual Report

(2013) the mostly occurred top diseases were: upper respiratory disease and diarrhea 35% and 9.8% respectively, which are as a result town households discharging there wastewater in to the environment without any treating mechanisms.

Heavy metals which might build up within the body over time square measure famed to cause varied health issues, such as; cancer, procreative and biological process disorders, neurologic problems; vas, skeletal, blood, system, excretory organ and nephritic problems; headaches; physiological reaction, nausea and diarrhoea; respiratory organ damage; contact dermatitis; and brittle hair and hair loss. Some are hormone disruptors while others are respiratory toxins. They can be ingested or absorbed through the skin, particularly broken skin. There are thirteen trace elements on the list of United State Environmental Protection Agency, EPA (2015) priority pollutants including antimony(Sb), arsenic(As), beryllium(Be), cadmium(Cd), Chronium(Cr), copper(Cu), lead(Pb), mercury(Hg), nickel(Ni), selenium(Se), silver(Ag), thallium(Th), and zinc(Zn).

Cadmium also found in body & hair creams and naturally in the environment are absorbed into the body through dermal contact (Ayenimo, Adeeyinwo, & Amoo, 2005) stored in the kidney and the liver, although it can be found in almost all adult tissues. It is considered to be “carcinogenic to humans” by the United States Department of Health and Human Services (Okereke, Ogidi, & Obasi, 2016). Ingestion of high levels of cadmium can lead to severe abdomen irritation, vomiting and diarrhoea, while exposure to lower levels for a long time can lead to excretory organ harm, bone deformity, and the ability of bones to break easily. Cadmium may interfere with metabolic process in plant, can bioaccumulate in aquatic organisms and enter the food chain (Adriano, 2001). Manifestation of toxicity in plants includes stunting chlorisis, necrosis and wilting, depressed photosynthesis and inhibits seed germination (Adriano, 2001). Cadmium is bio-persistent and once absorbed by an organism remains resident for several years. It’s presence in fish is a serious concern to human consumers.

Skin contact with lead occurs daily, and some have been found to be absorbed through the skin. Lead can be found in Lipstick via the use of contaminated raw materials or through the use of pigments that contain it (ATSDR, Agency of Toxic Substances and Disease Registry, 2007). The application of leaded eye powders (e.g., Surma, Kohl, Alkol) has been related to increased blood-lead levels in children and women (ATSDR, 2007). Pregnant women and young children are particularly vulnerable because it can cross the placenta with ease and enter the foetal brain. It can also be transferred to infants through breastfeeding and stored in bones. Lead exposure has also been linked to miscarriages, hormonal changes, reduced fertility in men and women, menstrual irregularities, delays in puberty onset in girls (Okereke, Ogidi & Obasi, 2016a). Lead and inorganic lead compounds have been classified as a suspected carcinogen to humans (ATSDR, 2007).

Mercury is also a typical ingredient found in skin-lightening soaps and creams. It is also found in other cosmetics, such as eye make-up, cleansing products and mascara (Okereke, Ogidi, & Obasi, 2016a). Mercury salts inhibit the formation of melanin, leading to lighter skin tone (Okereke, Ogidi & Obasi, 2016b). Mercury in cosmetics exists as inorganic (e.g. ammoniated mercury) and organic mercury compounds (ethyl mercury and phenyl mercuric salts). Inorganic mercury is employed in skin-lightening soaps and creams while organic mercury are used as cosmetic conservative in eye make-ups, mascara and cleansing products. The most adverse effect of the inorganic mercury contained in skin lightening soaps and creams is kidney damage (Okereke, Ogidi & Obasi, 2016c). Mercury in skin-lightening product may cause skin discoloration, skin rashes and reduction in the skin's resistance to microorganism infections. Mercury in creams, soaps and other cosmetic products will eventually be discharged into wastewater, enters the environment, methylated as methylmercury in fish and enters the food-chain. Pregnant women who consume fish containing methylmercury transfer the mercury to their foetuses, which can later result in neuro-developmental deficits in the children (Okereke, Ogidi & Obasi, 2016a). The possible effects of greywater constituents are presented in Table 2.2.

Table 2.2: Possible Effects of Sullages' Constituents

Constituent	**Measured Parameter**	**Possible Effects**
Lint, Solids, Particulates	Solids	Suspended matter can cause clogging of systems and shield microorganisms from disinfectants.
Organics	COD, BOD and TOC	Biofilm formation
Nutrients	Phosphorus, Nitrogen	Phosphorus and nitrogen are essential nutrients for plant growth. Excessive plant growth and nitrate build-up are not expected in properly sited, designed, and maintained systems.
Hydrogen ion concentration/ alkaline salts	pH/alkalinity	Effects of pH/alkalinity on soil and plants.
Salts	Total Dissolved Solids, Electrical Conductivity, Specific Elements (e.g., Na, Ca, Mg, Cl, B)	Excessive salt may damage some crops. Specific ions such as chloride, sodium, and boron at high concentrations may be toxic to some crops. Sodium may cause soil permeability problems.
Microorganisms	Total plate counts, Indicator organisms, Specific microorganisms	Fouling. Pathogenic bacteria, parasites and viruses are infectious agents of waterborne diseases.
Hypochlorite	And Combined Chlorine	Excessive amounts of free available chlorine (>0.05 mg/L) may cause leaf-tip burn and damage some sensitive crops. Some concerns about potential groundwater contamination by chlorinated organic.

2.4. AGRICULTURAL WASTES AS LOW COST ADSORBENTS IN TREATING WASTEWATER

Sullage has to be treated to get rid of substances which will be harmful to human health, to plants and soil, to get rid of substances which will be harmful to the environment and to remove substances that may clog the irrigation system.

Adsorption was coined in 1881 by the German scientist Henrick Kayser. Adsorption is a surface phenomenon which involves the adhesion of atoms, ions or molecules from gas, liquid or dissolved solid to a surface. This should not be confused with absorption in which a fluid (the absorbate) is dissolved by or permeates a liquid or solid (the absorbent), respectively. In adsorption technique, when a solution that contains adsorbable solutes is in contact with a solid with highly porous surface structure, liquid-solid intermolecular forces of attraction causes some of the solute from the solution to be concentrated or deposited at the solid surface (Arif *et al*., 2008). The solute retained (in the solid surface) in adsorption is called adsorbate, whereas the solid on which it is retained is called an adsorbent. Adsorption process is generally classified as physicosorption (characteristic of weak Van Dar Waals forces) or chemisorptions (characteristic of covalent donding) although the nature of the bonding depends on the details of the species involved. It may also occur due to electrostatic attraction.

Agricultural wastes or industrial by- product may be used as affordable adsorbents. Low- price adsorbent is outlined as any material that is abundant in nature, or is a by- product or waste material from another industry (Bailey *et al*., 1999). Adsorbent is called a low cost adsorbent if it needs very little process, is abundant in nature, or is a waste material or by-product from another industry (Nasin, Shaliza, & Piarapakaran, 2004). Agricultural waste otherwise called agro-waste can be animal waste (manure, animal carcasses), food processing waste, crop waste (corn stalks, sugarcane bagasse, drops and culls from fruits and vegetables, prunings). It is estimated that about 998 million tonnes of agricultural waste is produced yearly (Al-Baidhani & Al-Salily, 2016). The abundance and availability of agricultural wastes make them good sources of cheap raw materials for natural adsorbents (Daffalla, Mukhtar & Shaharun, 2010). Okereke, Ogidi & Obasi, 2016), in a study showed that banana peels are effective in removing inorganic anions in brewery effluent. Al-Baidhani & Al-Salily (2016) discussed the removal of heavy metals from wastewater using rice husk.

There is a great interest in the production of activated carbon from agricultural by-products (Rahmani *et al.*, 2009). Agricultural wastes such as cocoa pod husk (Rahman *et al.*, 2006), periwinkle shell (Aluyor & Badmus, 2008), walnut shell, peach stoner, physic nut waste, coconut shells, palm kernel shells, and bamboo stem wastes (Awoyale, Eloka-Eboka, & Odubiyi, 2012), have been used in the production of activated carbon thereby adding value to these agricultural wastes and thus, recycling them (Odubiyi, Awoyale & Eloka-Eboka, 2012). Eza *et al.* (2014) found activated carbon from coconut and oil palm shells as an effective anti-odour adsorbent on textile fabrics. Also, Odubiyi *et al*. (2012) showed that activated charcoal produced from cocoa pod husk great has potential for the elimination of heavy metal from wastewater. The adsorption of heavy metals (Pb and Cu) from effluent water has been investigated in batch condition. Activated carbon prepared from cocoa pod husk, carbonized at 500 ^{o}C and impregnated with 1.0 M HCl at 700^{o}C in a muffle furnace for 2 h, can adsorb heavy metals in effluent (wastewater) at varying pH, dosage and contact time. The maximum Pb removal efficiency of 78% at pH of 5.5 at a contact time of 1.9 h, and the maximum Cu removal from the effluent 97% at pH of 5.6 and a contact time of 2 h are in literature (Odubiyi *et al*., 2012). Study on the removal effects of organic pollutants in drinking water (42 species organic pollutants in 11 categories) by activated carbon, haydite and quartz sand with the method of solid-phase extraction (SPE) showed that the removal rates of organic pollutants by activated carbon, haydite and quartz have been put at 70.35%, 29.68% and 37.36%. Among all, activated carbon has been shown to be good in the removal of most organic pollutants (Mohamed, 2017).

2.5 ACTIVATED CARBON FORMS

Activated carbon is defined as carbon that has been heated or otherwise treated to increase its adsorptive capacity. During adsorption, activated carbon allows gases and chemicals to adhere to microscopic pores on the internal surface area of the carbon. "Activating" carbon is the process of making the carbon high in surface area to facilitate sorption. Adsorption opens a world of opportunity for industrial applications. Activated carbon is used to filter water, purify gas, and is even used as an ingredient in prescription medicines (Eze *et al.,* 2014).

The use of activated carbon to treat water was known over 2000 years ago, tracing back to 1500BC when Egyptian used charcoal as an adsorbent for medical purposes and purifying agent. During World War I, activated carbon was used in gas masks for protection against hazardous gases and vapors. Activated charcoal is formed from a variety of carbonaceous raw material including coal, coconut shells, wood, and other cellulosic materials. It has numerous applications in removing pollutants from air or water such as in spill cleanup, groundwater remediation, drinking water filtration, air purification and volatile organic compounds capture from painting, cleaning, fuel dispensing operations and alternative processes. Throughout early implementation of the 1974 Safe Drinking Act in the USA, EPA officials developed a rule that proposed required drinking water treatment systems to use granular activated carbon. Its usefulness derives from its large micropore (and sometimes mesopore), volume and high surface area (Okereke & Ogidi, 2016).

2.6 MECHANISM OF ACTIVATED CARBON PRODUCTION

There are two basic methods by which activation can be produce: physical activation and chemical activation.

Physical activation:

Physical activation comprises of two steps, carbonization of the precursor in an inert atmosphere and subsequent activation of the resulting char in the presence of carbon gasification reactants (gasous) such as carbon dioxide, steam or air at high temperature ranging between 800^0C and 1100^0C (Vis Wanatham, Indra & Varadarajan., 2009). In this process, pore creation and development is not equal in all parts. This process also takes longer time and consumes more energy for microporous activated carbon production. Hence, the yield is very poor.

Chemical activation:

Chemical activation involves heat treatment in an inert atmosphere at a temperature of 400-600^0C and then impregnation. The chemicals used as activators include transition metal salts like zinc chloride ($ZnCl_2$), acidic group such as phosphoric acid (H_3PO_4) and sulphuric acid (H_2SO_4) and alkaline group such as potassium hydroxide (KOH) and potassium carbonate (K_2CO_3). Compared to physical activation, chemical activation is more economical (uses lesser energy, short processing time and higher carbon yield) (Hidayu *et al.,* 2013; Sangchoon & Mokaya., 2015).

2.7 MECHANISM OF ADSORPTION

Adsorption of solute to the interior surfaces of adsorbent may take four steps (Aluyor & Badmus, 2008):

(1) Bulk transport: The adsorbates are transported to the boundary layer of water surrounding the adsorbent particle from the bulk solution. The transport is made possible by diffusion if the adsorbent is suspended in quiescent water, such as in a sedimentation basin, or by turbulent mixing, such as during turbulent flow through a packed bed of granular activated carbon (GAC) or when powdered activated carbon (PAC) is being mixed in a rapid mix or flocculator.

(2) Film transport: Adsorbates are transported by molecular diffusion through the stationary layer of water (hydrodynamic boundary layer) that surrounds adsorbent particles when water is flowing past them. The distance of transport and the time of this step are determined by the rate of flow past the particle; the higher the rate of flow, the shorter the distance.

(3) Intra-particle transport: After the adsorbates pass through the hydrodynamic boundary layer, they are transported through the adsorbent's pores to the available adsorption sites. Intra-particle transport may occur by molecular diffusion through the solution in the pores (pore diffusion), or by diffusion along the adsorbent surface (surface diffusion), then adsorption takes place.

(4) Adsorption of the solute on active sites: Once the absorbates reach an available site, the adsorption bond is formed between the adsorbate and adsorbent. This step is very rapid for physical adsorption and so one of the proceeding diffusion steps will control the rate at which molecules are removed from the solution. If a chemical reaction that changes the nature of the molecules takes place during the adsorption step, this chemical reaction may be slower than the diffusion steps and thereby controlling the rate of compound removal.

2.8 FACTORS AFFECTING ADSORPTION

The efficiency or adsorption capacity depends on the subsequent factors: (i) Nature of adsorbate (ii) Nature of adsorbent (iii) Specific area of the adsorbent (iv) pH (v) Activation of the adsorbent (vii) contact time.

The influence of those specific factors on the sorption mechanism will be described in the following section.

Nature of adsorbate

Physical adsorption is non-specific in nature, so every metals is adsorbed on the surface of any solid to a lesser or greater extent (Çetinkaya Dönmez *et al.*, 1999). However, easily ionized materials are adsorbed to a greater extent whereas low ionized materials are adsorbed to a lesser extent (Davis *et a*l., 2003). pH is the controlling factor for ionization.

Nature of biosorbent

Physisorption could be a general development that doesn't depend upon the character of adsorbent (solid). Chemosorption is restricted and depends on the character of each adsorbent and adsorbate (Naiya et al., 2009). As an example, nickel, platinum and iron readily adsorb hydrogen; silica gel adsorbs moisture, and tungsten adsorbs oxygen. The common adsorbents are biosorbents, carbon, activated carbon, metal oxides, silica gel, alumina and clay. Each adsorbent has its own characteristics, and functional groups are the main metal binding factors.

Specific area of the adsorbent

Specific area means the surface area available for adsorption of 1 gram of adsorbent. The amount of adsorption depends on the surface area of the solid. The more the surface area the more will be the biosorption (Vilar, Botelho & Boaventura, 2007a; Vilar, Botelho & Boaventura, 2007b). The pores of a porous adsorbent must be so large that the molecules of the adsorbate ions can enter through them (Peigney *et al*., 2001).

pH of water

The solubility of adsorbates like metals depends upon pH. On the other hand the activation of binding sites also depends on pH. For instance, hydrogen and hydroxide ions are adsorbed quite strongly; the adsorption of other ions is influenced by the pH of the solution. In general, adsorption of a typical organic pollutant from water is enhanced with decreasing pH. Metal adsorption largely depends on the nature of the species solution i.e pH. At lower hydrogen ion concentration, H+ competes with metals for the exchange sites within the system thereby partially releasing the latter. The heavy metals are completely released under circumstances of extreme acidic conditions (Annadurai *et al.,* 2003). As it is anticipated, at lower pH, more protons are available, thereby decreasing electrostatic attraction between cationic adsorbate and positively charged sites of adsorbent. This case may be the reason for the decrease of adsorption below pH 7.5. When the pH value of the solution is increased, the surfaces of the adsorbent become negatively charged. Additionally, the concentration of OH^- ion also increases with the increase of pH, and no exchangeable anions on the outer surface of the adsorbent will remain at higher pH values (Namasivayam, Sangeetha, Gunasekaran, 2007; Ozcan *et al*., 2005; Yu *et al*., 2001).

Activation of the adsorbent

The adsorption process solely depends on the activation of the outer surface of adsorbents and adsorbate. Activation is done by using some activation agents in the adsorption process.

Normally some acidic or alkaline solution or salt is used for activation. After adding activation agent, the ionizing strength and pH of the system changes and either adsorbent or adsorbate or both become activated (Aksu, 2001). As a result of this the adsorption capability becomes quicker and increase adsorption capacity.

Contact time

As reported in the literature, the metal sorption is often rapid, and usually complete in less than one hour (Gadd, 1988). Binding of metal ions to aquatic particulates is considered to be a fast chemical reaction, with an equilibrium time depending only on the mass transfer resistance (Waite *et al.*, 1994). To establish an appropriate contact time between the biosorbents and metallic ions solution, adsorption capacities of metal ion need to be measured as a function of time. Ordinarily the removal is higher within the starting and step by step drops with time. That is most likely because of the larger surface area of the biosorbents being available at the start for the adsorption of metals. Because the surface surface assimilation sites become exhausted, the uptake rate is controlled by the speed at that the adsorbate is transported from the outside to the inside sites of the adsorbent particles (McKay, 1982).

2.9 COLUMN ADSORPTION

There are two types of adsorption studies viz. batch mode and continuous flow mode or column studies. The batch operations are useful only for the treatment of small quantities of wastewater. Batch equilibrium experiment shows the adsorption capacity of the adsorbent, that provides elementary info concerning about the effectiveness of adsorbate–adsorbent system (Suksabye, Thiravetyan & Nakbanpote, 2008). The continuous flow system is an effective process for the treatment of large-scale wastewater volumes and cyclic adsorption/desorption. The experimental breakthrough curve is foretold from column adsorption studies which further helps in the determination of column bed operation life span and regeneration time. Additionally, basic engineering information is simply obtained from the continuous flow systems. In fact from industrial point of view, the removal of pollutants including dyes, heavy metal ions etc. using continuous flow systems is found to be very useful and reliable. Although batch laboratory adsorption studies provide useful information on the application of adsorption to the removal of specific waste constituents, continuous column studies provide the most practical application of this process in wastewater treatment. The reason for this is that the high adsorption capacities in equilibrium with the influent concentration rather than the effluent concentration can be achieved (Goel *et al.*, 2005). Several models are developed within the past to predict the sorption breakthrough behavior with high degree of accuracy. A number ofadsorbtion models are: The Bohart-Adams (1920), Thomas (1944) and Yoon-Nelson model (1984) (Goel *et al.*, 2005).

CHAPTER THREE

MATERIALS AND METHOD

3.1 MATERIALS

i. Agrowastes - Rice Husk, Coconut Husk, Corn Cob.

ii. Sullage from FUTO Male and Female Hostel.

Collection of Samples

Agrowastes were sourced from Relief Market, Owerri, Imo State. Sullage were collected from male and female hostels of Federal University of Technology, Owerri, in a sterile 10 litre container. All the analyses were carried out at Federal University of Technology, Owerri.

3.2 METHOD

3.2.1 Preparation of activated carbon from rice husk, coconut husk and corn cob.

Carbonization and activation of absorbent

The method of Grigis & El-Hendawy (2002) was adopted with slight modification as follows: 750 g of each agrowaste was washed with deionized water, dried and crushed using locally made grinder. They were carbonized in electric muffle furnace at 600^oC for 45 min. The carbonized or pyrolyzed sample was sieved using 1.18 mm mesh size. Activation was achieved by mixing the carbonized sample with 20% H_3PO_4 solution at a ratio of 1:1 (Acid: Char) and stirring for 30 min. After that, the sample was filtered, washed with deionized water until the washed off water gave a pH of 7.0. Washed activated carbon was then dried at 120^oC in an oven. The dried sample was stored in airtight plastic container.

Determination of the carbonization yield:

The carbonization yield was determined according to Swrap & Umesh (2015):

$$\text{Yield (\%)} = \frac{\text{Weight of carbon produced}}{\text{Weight of raw sample used}} \times 100$$

Adsorption column

The adsorption column was constructed according to the work of Swrap & Umesh (2015) but with little modifications. An apparatus (burrete) of 50 cm length and 1cm diameter, and a collection flask were provided for this experiment. An outlet was provided at the bottom of the tank. Experiment was conducted by placing the individual adsorbents in column apparatus separately at first and then in combination. From the tank waste water was allowed to pass through the apparatus and collected water analyzed. Experiment was carried out at room temperature of 30 ± 2^0 C.

3.3 CHARACTERIZATION OF WASTE WATER

3.3.1 Chemical Characterization (APHA, 2013).

pH

pH was determined by taking the pH readings for the before and after treatment of the effluent using pH meter. It was first calibrated with a buffer of 7.0.

Biochemical Oxygen Demand Analysis

The BOD_5 was determined using DO_2 meter. The DO_2 meter was calibrated using 5% sodium sulphate solution. The probe of the meter was inserted into the sample after which the meter switched on for about 10 minutes. The readings were recorded in mg/l. The sample was incubated in a 250 ml wrinkler's bottle for a period of 5days at 20^oC and the DO_2 of the fifth day recorded by inserting the probe again into the sample. The difference in the DO_2 (5) and DO_2 (1) was recorded and calculated as the BOD_5.

BOD_5 (mg/L) = DO_2 (5) – DO_2 (1)

Chemical Oxygen Demand (COD)

Some 20 ml of the sample plus blank was measured into a 250 ml conical flask. After, 10ml of dichromate solution ($K_2Cr_2O_7$) (0.25N) and 30 ml of concentrated sulphuric acid were added. Samples were cooled in a cold waterbath. The mixture was diluted to about 150 ml and 3 drops of ferroin indicator added. The mixture was titrated with ferrous ammonium sulphate solution (O.5N $FeSO_4$. $7H_20$) until there was a change in colour.

$$\text{COD, (mg/litre)} = \frac{(B - A)(N)(8000)}{V}$$

Where

A = volume of ferrous ammonium sulphate solution used for sample titration in ml

B = volume of ferrous ammonium sulphate solution used for blank titration in ml

N = normality of the ferrous ammonium sulphate solution

V = sample volume in ml

Chloride (Argentometric Method)

Measured 50ml of the wastewater sample was transferred into conical flask and 3-4 drops of indicator 5% potassium chromate indicator added. The solution in the conical flask was titrated with 0.02N silver nitrate to a reddidh brown end point using a microburette. A blank titration was done as above using deionised water.

$$\text{Chloride, (mg/l)} = \frac{(V_1 - V_2) \times N \times 35.5 \times 1000}{\text{Volume of sample}}$$

Where

V_1 = volume of sample titer

V_2 = volume of blank titer

N = normality of silver nitrate used (0.02N)

Phosphate (Ascorbic Acid Method)

Ammonium heptamolybdate (6.0 g) was weighed and dissolved in 150 ml distilled water in 250ml conical flask. Ascorbic acid (2.6 g) was dissolved in 50 ml of distilled water in 1litre volumentric flask to give 0.0007 M. Then, 0.4 g of disodium ETDA was dissolved in the 0.0007M ascorbic acid and O.14 g of potassium antimony tartrate weighed and dissolved in 20 ml distilled water (0.0000086 M). Stock of concentrated sulfuric acid (1M) was prepared by dissolving 10ml of the stock in 50ml distilled water.

Phosphate stock (1000 mg/L PO_4^{3-}) was prepared by weighing accurately 1.532 g of potassium phosphate trihydrate in 250 ml of distilled water. Thereafter, 0.5, 1.5, 2.0, and 2.5 ppm PO_4 were prepared by proper dilution with distilled water for calibration curve.

Measured volume (12.4 ml) of the ammonium molybdate solution was transferred into a 50 ml volumentric flask and then 10 ml sulfuric acid was added, swirled, and 2.3 ml of antimony potassium tartrate added. The mixture was swirled properly to mix and the mixture made up to the mark with distilled water.

Spectrophotometric determination was carried out by adding 0.4 ml molybdate reagent to 20ml of standard or sample in a test tube and swirled to mix. Also, 0.4 ml of L-ascorbic acid was added and swirled. The light absorption of the solution was measured at 820 nm wavelength using spectrophotometer.

Nitrate, (NO_3^-) determination

To each of standard nitrate solution of different concentrations were added 10 ml of H_2SO_4 and swirled. The beakers containing the solutions were allowed to reach thermal equilibrium inside a cold water bath prior to heating. To each of the beakers was added 0.5 ml of brucine-sulphahilic acid reagent, swirled properly to mix and placed in 100^oC water bath for 25 minutes. After heating, the beakers were removed from the hot water bath, immersed in a cold water bath and allowed to reach thermal equilibrium of 20-25^oC. The mixtures in the beakers were hence analyzed with spectrophotometer and the absorbance read at 410 nm wavelength. The samples and blank were treated in the same manner.

Sulphate (Turbidometric Method)

Deionised water (100 ml) was poured into 50 ml of buffer solution, and then transferred into 125 ml Erlenmeyer flask containing a clean magnetic stirring bar. Some 10 ml of deionised water, 6 ml of buffer reagent, and 10 ml of the standard solution were added into the flask. It was gently swirled to mix. 0.2 g of $BaCl_2$ was added to the flask, placed on the magnetic stirrer and stirred for 58 to 62 seconds. The solution was allowed to stand undisturbed for 2 minutes. The light absorption of the solution was measured at 420 nm wavelength using spectrophotometer.

3.3.2 Heavy Metal Concentrations

Heavy metal analysis was conducted using Agilent FS240AA Atomic Spectrophotometer according to the method of APHA (2005). Atomic absorption spectrophotometer's working principle is based on the sample being aspirated into the flame and atomized when the AAS's light is directed through the flame into the monochromator, and unto the detector that measures the amount of light absorbed by the atomized element in the flame. Since the metal have their own characteristic absorption wavelength, a source lamp composed of the element is used, making the method relatively free from spectral or radiational interferences. The number of energy of the characteristic wavelength absorbed within the flame is proportional to the concentration of the element in the sample.

3.3.3. MICROBIAL ANAYLSIS

Preparation of media and diluents

Bacteriological (nutrient agar, mannitol salt agar, eosin methylene blue agar and salmonella shigella agar) and mycological (potato dextrose agar) media were prepared according to manufacturer's specification.

Physiological saline used as diluents was prepared by dissolving 9.8 g of sodium chloride in 1000 ml of distilled and dispensed in 9 ml portions. Both diluents and media were sterilized in an autoclave at 121^0C for 15 mins (Braide, Offor-Emenike & Oranusi, 2014; Cheesbrough, 2000).

Preparation of samples and inoculation

One milliliter of the samples (wastewater) was transferred into 9 ml of the diluents and serially diluted until 10^6 were obtained. Aliquot portion (0.1 ml) of appropriate dilution was inoculated into the pre-sterilized and surface dried nutrient agar medium. Inocula were spread evenly to ensure uniform and countable colonies. Plates were incubated at 28^oC temperature 24 - 48 hours (Cheesbrough, 2000).

Determination of microbial population

Colony counts obtained on the media were expressed as colony forming units per ml (CFU/ml) by multiplying with dilution factor to obtain total population (Braide *et al.*, 2014).

Characterization and identification of microbial isolates

Microbial isolates were characterized based on cultural (colonial), microscopic and biochemical methods and isolates identities matched with reference to standard manuals for the identification of bacteria (Braide *et al.*, 2014; Cheesbrough, 2000).

Characterization of microbial isolates

Microscopic characterization

Gram staining test

An isolate smear was developed with a drop of water on grease free glass slide and permitted to dry. The smear was mounted by delicate heating and flooded with crystal violet, then allowed to stand for 30 seconds and rinsed with water. Lugol's iodine was then added, allowed to stand for 30 seconds and rinsed with water and acid alcohol, till no colour change is observed. Safranin was used to counter stained, allowed to stand for 10 seconds, rinsed with water and air dried. On the slide, oil immersion (1 drop) was added and viewed using the microscope's x100 objective lens (Braide et al., 2014).

Spore staining test

This test was used to verify the presence of spores. Isolates were heat fixed on a slide and flooded with 5% malachite green. It was steamed for 3 minutes (without boiling), allowed to dry, washed off and later stained with Safranin for 30 seconds. The slide then rinsed was dried with filter paper was observed under the microscope using oil immersion lens. Green colour indicates spore while colour pink indicates negative spores.

Motility test

To determine the motility of bacteria isolated. A semi-solid agar medium was used. The medium was poured into test tubes, autoclaved and allowed to set in an upright position. Isolates were inoculated using an inoculation needle by stabbing it into the medium in the test tube and then incubated at 37°C for 24 hours. Diffused growth from the straight line of inoculation indicates positive result (Cheesbrough, 2000).

Biochemical characterization of bacteria isolates

Microorganisms were further subjected to few biochemical tests (Braide *et al.,* 2014; Cheesbrough, 2000).

Catalase test

This test demonstrates the presence of catalase, an enzyme that catalyses the release of oxygen from hydrogen peroxide. One drop of 3% hydrogen peroxide solution was placed on a clean slide. A loop full from 24 h culture was added. The release of bubbles (of oxygen) indicated the presence of catalase in the culture under test.

Coagulase test

The coagulase test identifies whether an organism produces exoenzyme. This enzyme clots the blood plasma by a mechanism that is similar to normal clotting. *Staphylococcus aureus* is the only significant disease causing bacteria of humans that produce coagulase. Thus coagulase test is a good indicator of *S. aureus.*

Sample were added to rabbit plasma in a test tube and incubated at 37^0C for a specified period of time. Presence of clot (coagulation) within four hours indicates positive result and indicates a virulent *S. aureus* strain. After 24 hours, the absence of coagulation indicates a negative result.

Oxidase test

This was used to check for the presence of terminal enzyme cytochrome c oxidase or cytochrome a. The presence of cytochrome c oxidase in the organisms is determined by the reagent tetramethyyl-p-phenylene diamine dihydrochloride. It is used for the identification of *Pseudomonas* spp, *Aeromonas* spp, *Neisseria* spp, *Vibro* spp and *Pasteurella* spp, all of which produce oxidase. Members of entrobacteriaceae give negative oxidase test.

A piece of paper was placed in a clean petri dish and 2-3 drops of freshly prepared oxidase reagent (1% tetramethyl-p- phenylene diamine dihydrochloride) was added. A small portion of culture was placed in the filter paper with the help of a sterile glass rod and a smear was made. Colour change was examined within 10 secounds. Blue-purple colour indicated the presence of terminal enzyme cytochrome c oxidase or cytochrome a.

Oxidation / Sugar fermentation

This test was used to differentiate between bacteria groups that oxidize carbohydrate such as members of Enterobacteriaceae. One milliliter (1 ml) of 10% glucose, maltose, lactose, fructose, mannitol, and sucrose were separately transferred into duplicate tubes containing 9ml of sterile Hugh and Leifson's medium to obtain a final concentration of 1% of each of sugar. The tubes were stab-inoculated in duplicates while two uninoculated tubes served as control. One set of the duplicate tubes were covered with Vaseline and one control to discourage oxidative utilization of sugar. All tubes were incubated at 37^{o}C for 48 h and observed for acid production in the culture. Yellow colouration in the open tubes indicated acid production, suggesting oxidative utilization of the sugar. Acid production in the sealed tubes indicated a fermentative reaction.

Hydrogen sulphide production (H_2S) test

Isolates were inoculated into a tube containing triple sugar iron agar by stabbing the agar to the bottom and streaking the surface of the slant. The inoculated tube was incubated at 37^{o}C for 72 h and was examined daily. Black precipitate indicated H_2S production while yellow colouration indicated lactose, glucose and sucrose fermentation.

Urease test

Isolates were inoculated in Urease Agar slant in McCartney bottle at 30^{o}C for 4 hours and then overnight. Pink colour in the medium indicated a positive result.

IMViC TEST

This test determined the physiological properties of microorganism. It consists of four different tests: indole test, methyl-red test, Voges Proskaeur test and citrate utilization test. They are especially useful in the differentiation of Gram-negative intestinal bacilli, particularly *Escherichia coli* and the *Enterobacter-Klebsiella* group.

Indole test

This test demonstrates the ability of certain bacteria to decompose the amino acid-Tryptophan to Indole. The bacteria isolates were inoculated into the medium and incubated at 37°C for 48 hours. At the end of incubation period, 3 drops of Kovac's reagents was added and then shaken. A red colour ring at the interface of the medium indicates a positive result.

Methyl red test and Voges-Proskaeur

Methyl red and Voges-Proskaeur test are considered together since they are physiologically related. Opposite test is usually obtained from the MR and VP test, that is, MR+, VP-, or MR-, VP+.

Methyl red test demonstrates the capacity of different organisms to produce acid from the fermentation of sugar (dextrose).

Inoculated glucose phosphate medium was incubated at 37°C for 2 days. Two drops of methyl red solution were added shaken well and examined. Red colour indicated positive reaction while yellow indicated negative reaction.

The Voges-Proskaeur test demonstrates the ability of organisms to produce acetoin from glucose metabolism. One milliliter (1 ml) of six percent alcoholic solution of alpha-naphtol and 1ml of 16% KOH was added into 1ml of the culture and stood for 15-20 minutes. Red to pink colour indicates a positive test.

Citrate utilization test

This test demonstrates the ability of an organism to use citrate as its only source of carbon. It is effective in used to assist in the identification of Enterobacteria. It was carried out using Simmon's citrate agar.

The slopes of the media were prepared in bijou bottles as recommended by the manufacturers. A sterile straight wire was used to the slope with a saline suspension of the test organisms before stabbing the butt. The bottles were incubated at 35ºC for 48 h. Bright blue colours in the medium indicates positive test while no change in colour indicates negative citrate test.

3.4 EFFICENCY OF PERCENTAGE REMOVAL

The efficiency of percentage removal was calculated according to Ahmad *et al*., 2016.

$$\text{Removal efficiency} = \frac{C_o - C_k}{C_o} X\ 100$$

Where

C_o = Initial concentration

C_k = concentration after treatment

3.5 STATISTICAL ANALYSIS

Analysis of Variance (ANOVA) and Student t-Test at 0.05 (95%) significant level were used to analyze data generated.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 RESULTS

Percentage Carbon Yield

The percentage yield after carbonization shows that rice had a higher carbon yield followed by corn cob and coconut husk as shown in Figure 4.1. Carbon yield (%) for rice husk was 35.7%; corn cob 22.0% and coconut husk 10.0%.

Chemical Parameters of Untreated Sullage

In Table 4.1, COD mean values ranged from 342.2±71 mg/l - 482.2±87 mg/l while BOD mean values ranged from 1.23.2±25 mg/l - 175.0±31 mg/l. pH mean values ranged from 7.01±0.1 - 7.33±0.1. Phosphate had a range of 4.25±1.3 mg/l - 8.77±1.7 mg/l. Chloride had a range of 21.9±4.4 mg/l - 31.0±5.5 mg/l. Nitrate had a range of 7.74±9.5 ppm - 28.6±24 ppm while sulphate had a range of 289.51±21.7 ppm -347.08±67.1 ppm. At $p<0.05$, there was no significant difference between male, female and combined hostels sullage, although the parameters were higher in the samples from male sullage.

Concentration levels of Heavy Metals in untreated sullage

From Fig 4.1, there were no significant difference in the sullage at $p<0.05$ for the four parameters. Mercury (Hg) mean value ranged from 2.66±1 ppm - 4.01±0.6 ppm. Chromium (Cr) ranged from 0.015±0.1 - 0.14 ppm, Cadmium (Cd) ranged from 0.071±0.1 - 0.076±0 ppm while lead (Pb) ranged from 0 - 0.202 ppm. Apart from mercury, the heavy metals parameters were higher in male sullage.

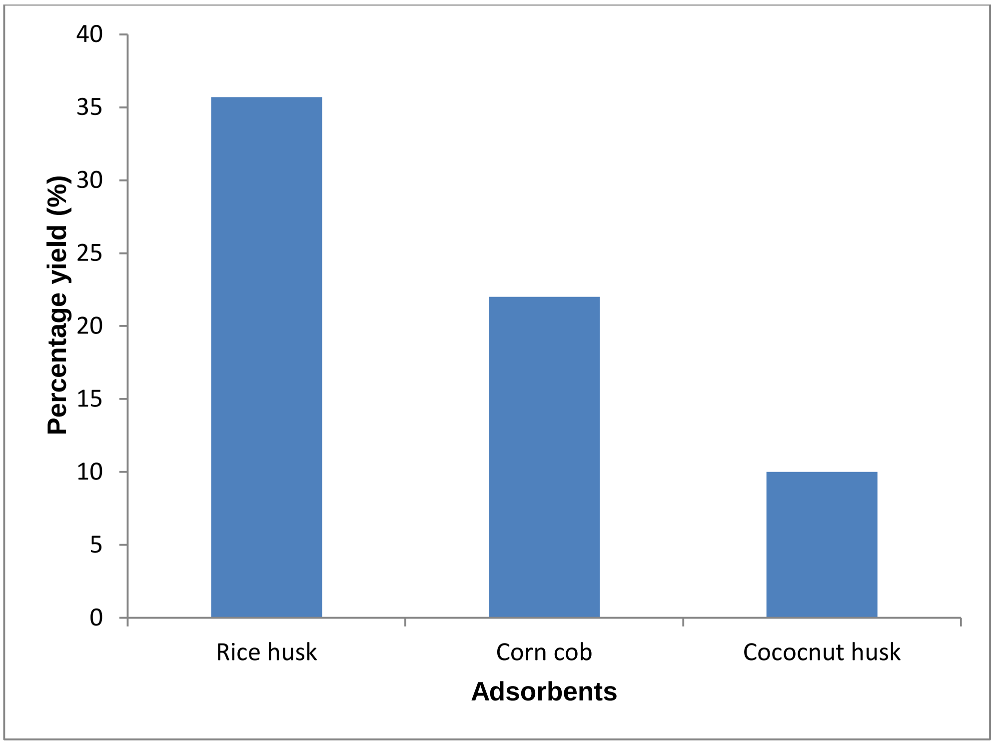

Fig 4.1: Percentage carbon yields of agrowastes

Table 4.1: Chemical properties of untreated sullage.

Heavy metals		Samples		EPA	WHO
	M	F	M F	limits	limits
COD (mg/l)	486.2±87.0	342.2±71.0	395.5±64.0	250	200
BOD (mg/l)	175.0±31.0	123.2±25.0	142.4±23.0	50	40
pH	7.33±0.1	7.01±0.1	7.27±0.1	6-9	6.5-8.5
PO_4^-(mg/l)	7.68±3.2	4.25±1.3	8.77±1.7	10	10
Cl^- (mg/l)	31.0±5.5	24.9±3.9	21.9±4.4	250	200
NO_3^-(mg/l)	28.6±24	7.74±9.5	13.83±11.8	50	45
SO_4^{2-}(mg/l)	347.08±67.1	319.58±24.6	289.51±21.7	200	200

All values were expressed as Mean±SEM (Standard Error of Mean).

M - sullage from male hostel; F - sullage from female hostel; MF – combined sullage from male and female hostels. EPA Limits= Permissible limits; ND= Not Dtected

Table 4.2: Concentration levels of heavy metals in untreated sullage.

Heavy metals		Samples		EPA	WHO limits
	M	**F**	**M F**	**limits**	
Hg(ppm)	2.66±0.1	4.01±0.6	2.68±0.3	0.001	0.001
Cr(ppm)	0.14±0.3	0.115±0.0	0.13±0.3	0.05	0.05
Cd(ppm)	0.071±0.1	0.071±0.1	0.076±0.0	0.003	0.01
Pb (ppm)	0.2±0.1	ND	0.06±0.0	0.01	0.1

All values were expressed as Mean±SEM (Standard Error of Mean). M - sullage from male hostel; F - sullage from female hostel; MF – combined sullage from male and female hostels. ND= Not Dtected

Microbial load of untreated sullage.

From Table 4.3., Total Heterotrophic Count (THC) ranged from $6.9x10^{10}$ - $1.2x10^{11}$ cfu/ml; Total Coliform Count (TCC) ranged from $3.7x10^{6}$ - $6.4x10^{6}$ cfu/ml while Total Fungi Count (TFC) ranged from $1.5x10^{8}$ - $2.2x10^{10}$ cfu/ml. At $p<0.05$, there was no significant difference between male, female and combined hostels sullage.

Biochemical Characteristics of Bacterial Isolates

From Table 4.4, the bacteria genera identified include *Micrococcus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp and *Bacillus* sp.

Colonial and microscopic characteristics of fungal isolates

Saccharomyces sp & *Penicillium* sp were the only fungi found in the sullage as shown in Table 4.5.

Table 4.3: Microbial load of untreated sullage.

Parameter		Samples		WHO
(CFU/ML)	M	F	M F	Limits
THC	$6.9x10^{10}$	$1.2x10^{11}$	$8.9x10^{10}$	$\leq 1.0x10^3$
TCC	$4.9x10^6$	$6.4x10^6$	$3.7x10^6$	$\leq 1.0x10^2$
TFC	$1.5x10^8$	$2.2x10^{10}$	$1.69x10^{10}$	-

All values were expressed as Mean.

M - sullage from male hostel; F - sullage from female hostel; MF – combined sullage from male and female.

THC= Total Heterotrophic Count; TCC= Total Coliform Count; TFC= Total Fungi Count

Table 4.4: Biochemical Characteristics of Bacterial Isolates from sullage.

Gr	Mo	Cat	Oxi	Coag	In	MR	VP	Cit	Ure	H_2S	G	S	L	Identity of isolate
+	-	+	-	+	-	-	+	-	+	-	+	+	+	*Staphylococcus aureus*
+	-	+	-	-	-	+	-	+	+	-	-	-	-	*Micrococcus luteus*
+	+	+	-	-	-	-	+	+	-	-	+	-	-	*Bacillus* sp
-	+	+	-	-	-	+	-	+	-	+	+	-	-	*Salmonella* sp
-	-	-	-	-	-	+	-	-	-	-	-	-	-	*Shigella* sp
-	-	+	--	-	-	+	-	+	-	-	+	+	+	*Enterobacter* sp

Gr, Gram Reaction; Mo, Motility; Cat, catalase; Oxi, oxidase; Coag, coagulase; In, indole; MR, Methyl Red; VP, Voges Proskaeur; Cit, citrate, Ure, urease; H_2S, hydrogen sulfide production, G, glucose, S, sucrose; L, lactose; M, maltose.

Table 4.5: Colonial and Microscopic Characteristics of Fungal Isolates

Colonial characteristics	Microscopic characteristics	Identity of isolates
Small circular moist and shiny yellow colonies	Gram positive spherical budding cells	*Saccharomyces* sp
Small circular moist and shiny cream colonies	Large Gram positive oval budding cells	*Saccharomyces* sp
Cream butyrous raised dull and mucoid colonies	Gram positive ellipsoidal budding cells	*Saccharomyces* sp
Dirty green powdery spores enclosed in white mycelium	Septate hyphae, conidia mob like	*Penicillium notatum*

Chemical parameters of sullage treated with absorbents.

From Table 4.6, there was a reduction in the chemical parameters from the control parameter (MF). COD ranged from 122.4 to 190.5 mg/l; BOD ranged from 44 – 68 mg/l; pH, 7.07 – 7.17; PO_4^-, 3.61 – 6.63 ppm; Cl^-, 17.28 – 21.13 mg/l; NO_3^-, 0 – 12.79 ppm and sulphate 117.7 – 251.7 ppm. Activated carbon from rice husk + corn cob + coconut husk gave the best BOD_5 treatment (44.0mg/l) while corn cob gave the least treatment (190.5). Activated Carbon from Rice husk + corn cob + coconut husk (122.4 mg/l) gave the COD best treatment while corn cob gave the least COD. For sulphate, activated carbon from rice husk + corn cob + coconut husk (117.70 ppm) and rice husk (147.8 ppm) performance better while corncob (175.26 ppm), coconut husk (251.72 ppm) had the least performance.

Concentration levels of Heavy Metals in sullage treated with adsorbents.

From Table 4.7, there was a reduction in the chemical parameters from the control parameter (MF). All the adsorbents completely removed chromium. All the adsorbents completely removed cadmium except activated carbon from corn cob+coconut husk and rice husk +corn cob +coconut husk. Activated carbon from rice husk, corn cob, and rice husk+corn cob completely removed lead. Mercury was the least to be removed by the adsorbents. Mercury was removed by the adsorbents in this order, corn cob+coconut husk, rice husk, rice husk +corn cob +coconut husk, coconut husk, rice husk+corn cob, rice husk+ coconut husk and corn cob.

Table 4.6: Chemical properties of sullage treated with adsorbents

Parameters	Adsorbents								
	Rh	Cc	Ch	Rh +Ch	Rh + Cc	Cc +Ch	Rh +Cc + Ch	EPA limits	WHO limits
COD(mg/l)	158.7±21	190.5±25	188.1±15	170.1±19	127.8±30	187.7±31	122.4±10*	250	200
BOD(mg/l)	57.1±76*	68.6±9.1	67.7±5.2	46.0±11*	61.3±6.7	67.6±11.3	44.0±3.5*	50	40
pH	7.15±0.1	7.18±0.1	7.18±0.1	7.12±0.1	7.13±0.1	7.13±0.1	7.08±0.1*	6-9	6.5-8.5
CL^- (mg/l)	20.5±5.5	17.3±3.3	20.3±4.0	18.5±3.1	20.9±4.6	17.8±2.4	21.1±4.4	250	200
PO_4^-(ppm)	3.61±0.4*	5.61±1.6	5.34±1.9	4.88±2.9	6.60±0.7	6.30±1.1	3.89±05	10	10
NO_3^-(ppm)	8.8±10.7	12.5±12.7	4.1±3.2	12.8±14	ND	11.1±13.3	ND	50	45
SO_4^2(ppm)	147.8±7.5*	175.3±23.3	251.7±22.3	163.2±1.7*	178.7±9.7	183.9±31.2	117.7±25.3*	200	200

All values were expressed as Mean±SEM (Standard Error of Mean). MF= combined untreated sullage from male and female. Rh=rice husk; Cc=corn cob; Ch=coconut husk; Rh+Ch= rice husk+coconuthusk; Rh+Cc=rice husk+corncob; Cc+Ch=corncob+coconuthusk; Rh+Cc+Ch=rice husk +corn cob +coconut husk. LSD *, shows the value with significant different compared with MF at p≤0.05.

Table 4.7: Concentration levels of Heavy Metals in sullage treated with adsorbents.

Parameters **Adsorbents**

Heavy metals	**Adsorbents**							**FEPA limits**	**WHO limits**
	Rh	**Cc**	**Ch**	**Rh + Ch**	**Rh + Cc**	**Cc + Ch**	**Rh +Cc + Ch**		
Hg(ppm)	1.71±0.6	2.51±0.3	2.18±0.1	2.27±0.3	2.25±0.2	1.60±0.5	2.16±0.4	0.001	0.001
Cr(ppm)	ND	ND	ND	ND	ND	ND	ND	0.05	0.05
Cd(ppm)	ND	ND	ND	ND	0.001±0	ND	0.008±0	0.003	0.01
Pb(ppm)	ND	ND	0.06±0	0.06±0	ND	0.03±0	0.02±0	0.01	0.1

Rh=rice husk; Cc=corn cob; Ch=coconut husk; Rh+Ch= rice husk+ coconut husk; Rh+Cc= rice husk+corn cob; Cc+Ch= corn cob+coconut husk; Rh+Cc+Ch=rice husk +corn cob +coconut husk. MF= combined untreated sullage from male and female. All values were expressed as Mean±SEM (Standard Error of Mean).

Microbial load of sullage treated with adsorbents.

Table 4.8 shows that Rice husk+ corn cob+ coconut husk was more effective in inhibiting growth of microorganisms. This was followed by corn cob, rice husk+corn cob, rice husk+ coconut husk for Total Heterotrophic Count, corn cob+coconut husk, rice husk and lastly coconut husk. Total Heterotrophic Count after the treatment ranged from 1.69×10^{9} to 7.6×10^{10} cfu/ml; Total Coliform Count ranged from 2.2×10^{5} - 7.3×10^{8} cfu/ml while Total Fungi Count ranged from 1.0×10^{8} - 1.2×10^{9} cfu/ml.

Distribution of Bacteria and Fungi in sullage samples

From Table 4.9, *Bacillus* sp was observed to be present in the female hostel sullage only while *Micrococcus* sp was observed in the male hostel sullage. *Bacillus* sp was not found after the treatment with the activated agrowastes. *Saccharomyces* sp was observed to be present in the sullage even after the treatment with the adsorbents. *Penicillium* sp was observed to be present after the treatment with activated carbon from Coconut husk, and Rice husk + Corn cob + Coconut husk

Table 4.8: Microbial load of sullage treated with adsorbents.

Media	MF	Rh	Cc	Ch	Rh + Ch	Rh + Cc	Cc + Ch	Rh + Cc + Ch	WHO Limits
THC	$8.9x10^{10}$	$5.3x10^{10}$	$7.3x10^{9}$	$7.6x10^{10}$	$3.6x10^{10}$	$1.9x10^{10}$	$3.98x10^{10}$	$1.6x10^{9}$*	$\leq 1.0x10^{3}$
TCC	$3.7x10^{6}$	$2.3x10^{6}$	$7.3x10^{8}$	$1.1x10^{6}$	$3.9x10^{5}$	$3.6x10^{5}$	$2.2x10^{5}$	$6.5x10^{5}$	$\leq 10x10^{2}$
TFC	$1.69x10^{10}$	$1.67x10^{8}$	$7.3x10^{8}$	$1.3x10^{8}$	$1.0x10^{8}$	$2.6x10^{8}$	$1.2x10^{9}$	$2.0x10^{8}$	-

All values were expressed as Mean (CFU/ML). LSD *, shows the value with significant different compared with MF at $p \leq 0.05$.

Rh=rice husk; Cc=corn cob; Ch=coconut husk; Rh+Ch= rice husk+ coconut husk; Rh+Cc= rice husk+corn cob; Cc+Ch= corn cob+coconut husk; Rh+Cc+Ch=rice husk +corn cob +coconut husk. THC= Total Heterotrophic Count; TCC= Total Coliform Count; TFC= Total Fungi Count. MF= combined untreated sullage from male and female.

Table 4.9: General Distribution of Bacteria and Fungi in sullage samples

Samples	Bacterial isolates	Fungal isolates
Male	*Micrococcus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Female	*Bacillus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Male + Female	*Micrococcus* sp, *Bacillus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Rice husk	*Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Corn cob	*Enterobacter* sp, *Staphylococcus* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Coconut husk	*Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp, *Penicillium* sp
Corncob + Coconut husk	*Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Rice husk + Corn Cob	*Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Rice husk + Coconut husk	*Enterobacter* sp *Salmonella* sp, *Salmonella* sp, *Shigella* sp	*Saccharomyces* sp
Rice husk + Corn cob + Coconut husk	*Staphylococcus* sp, *Enterobacter* sp, *Shigella* sp	*Saccharomyces* sp, *Penicillium* sp

Efficiency removal of chemicals properties from sullage.

From Fig 4.2, COD percentage removal ranged from 51.8 – 69.05% and BOD, 51.8 – 69.09%. Nitrate percentage removal ranged from 7.5 - 100%, Chlorine 4.3% - 21.1%, phosphate, 8.67 – 50.3% and sodium 38.3% - 59.3%. The efficiency of removal was in this order: Rice husk + corn cob +coconut husk > rice husk + Corn cob > rice husk > rice husk + coconut husk > corn cob > coconut husk > corn cob+ coconut husk and rice husk + coconut husk. For their individual strengths, activated carbon performed in this order: rice husk > corn cob > coconut husk . In combination, rice husk + corn cob + coconut husk > rice husk + corn cob > corn cob + coconut husk > rice husk + coconut husk .

Efficiency of adsorbents in removal of heavy metals from sullage.

Fig 4.3 shows that chromium (Cr) was more removed by the adsorbents, followed by cadmium (Cd), lead (Pb) and mercury (Hg). Cr was 100% removed. Cd percentage removal ranged from 89.47 - 100%, Pb ranged from 0 - 100% while mercury was 6.24 – 40.2%. Efficiency removal was in this order: rice husk > rice husk + corn cob > corn cob > coconut husk > corn cob + coconut husk > rice husk + corn cob + coconut husk > rice husk + coconut husk. For their individual strengths, activated carbon performed in this order: rice husk > corn cob > coconut husk. In combination, rice husk + corn cob > corn cob + coconut husk > rice husk + corn cob + coconut husk > rice husk + coconut husk.

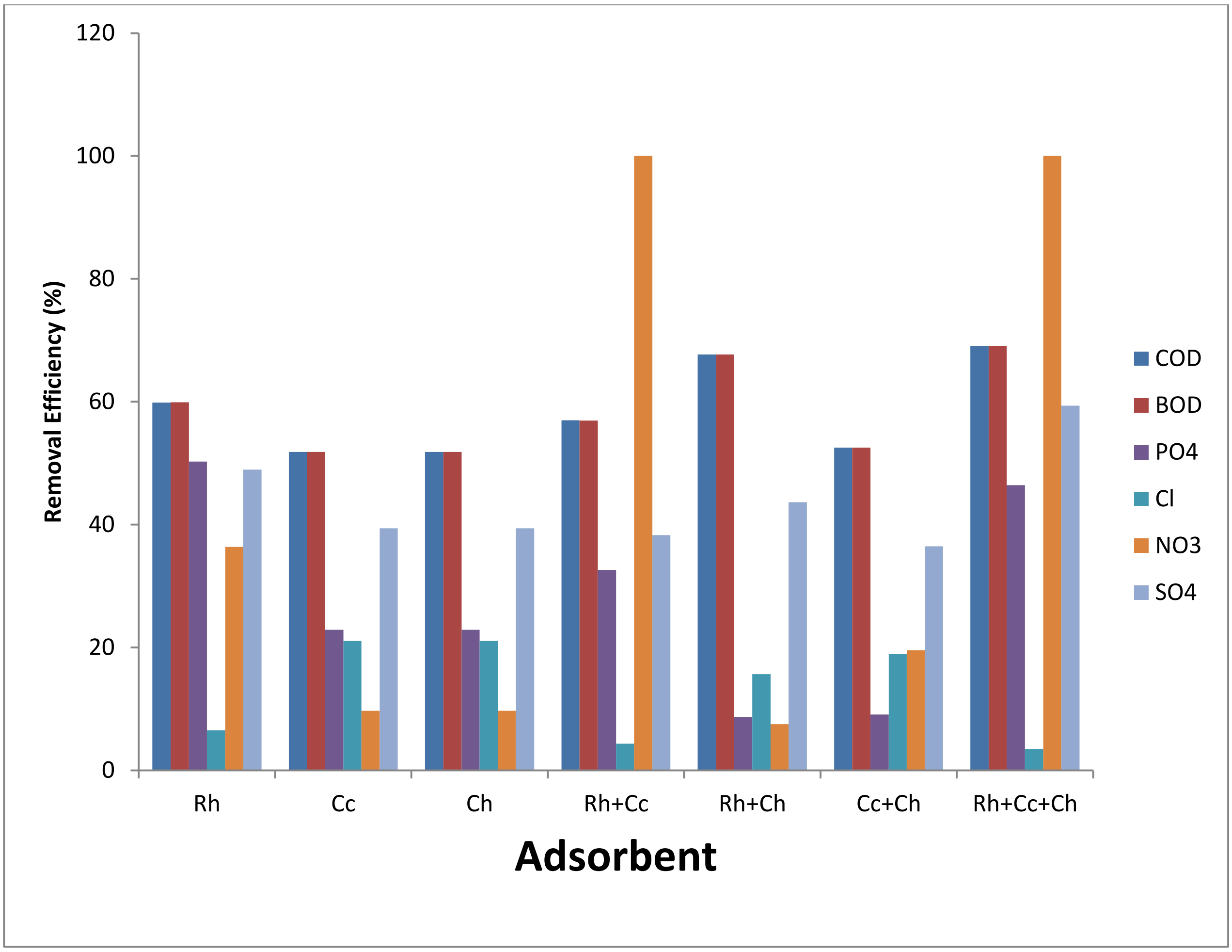

Fig 4.2: Efficiency of the adsorbents in the removal of chemicals properties from sullage.

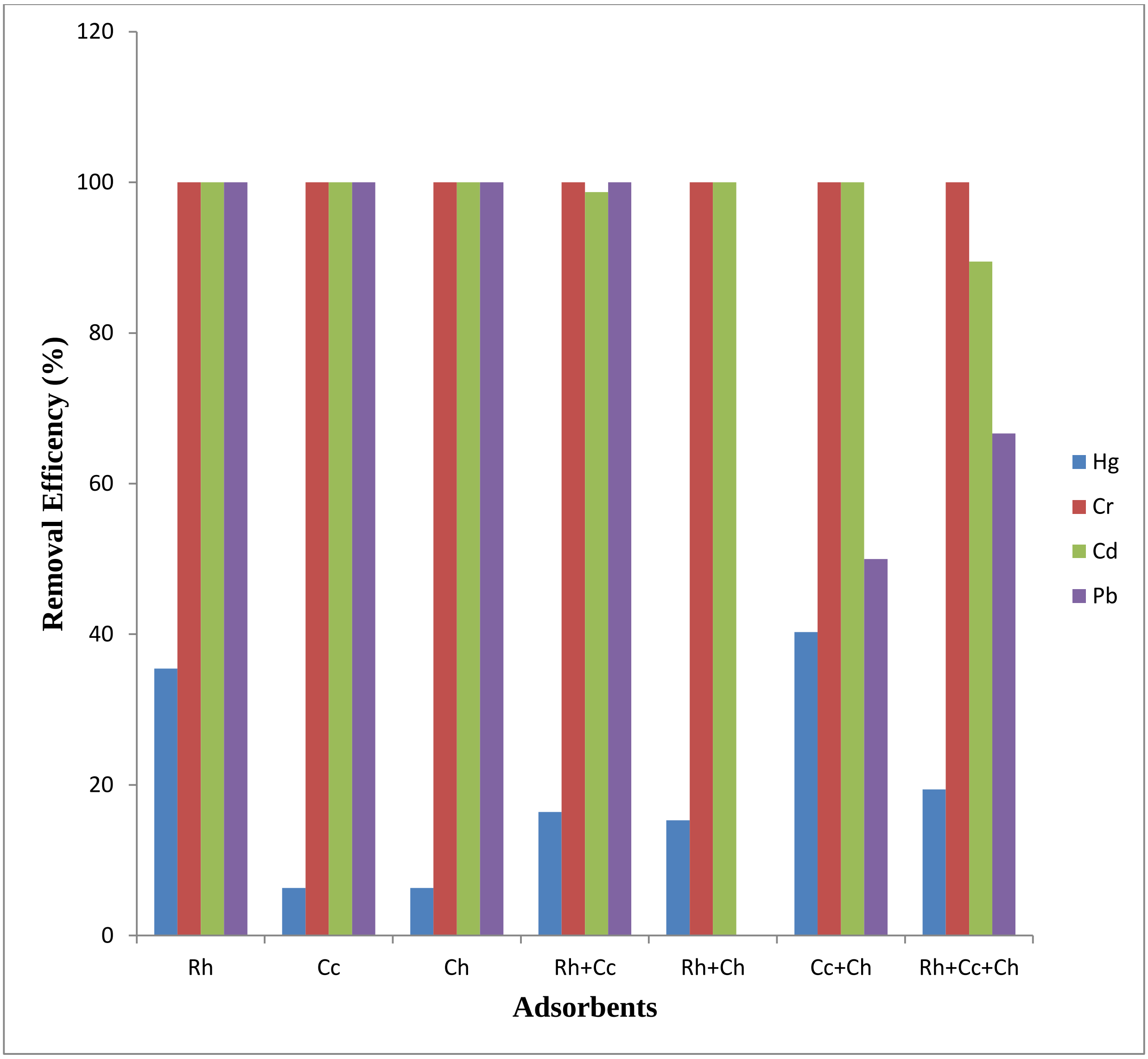

Fig 4.3: Efficiency of the adsorbents in the removal of heavy metals from sullage.

4.2 DISCUSSION

Percentage carbon yields of agrowastes

From the study, it was observed that rice husk had the highest percentage yield of 35.7% when compared to corn cob (22%) and coconut husk (10%) following treatment at the temperature of 600^0C. Abugu, Okoye, Ajiwe, Omuku & Umeobika (2015) reported that the yield loss is usually 60-70% as some CO_2 is volatilized. According to Abugu *et al.* (2015), increase in activation temperature above 700^oC leads to decrease in surface area and porosity of activated carbon. The carbonized agrowastes were chemically activated with phosphoric acid (H_3PO_4) before using them to treat the sullage. Activation process helps in fine pore structure formation. These pores are where adsorptive molecules are attracted. According to Singh, Abdullah & Chhotu (2018) chemical treatment, impregation ratio, temperature variation and yield, can enhance the pore structure of carbonized agrowastes.

Chemical properties of untreated sullage from hostels

From Table 4.1, apart from pH, phosphate, chloride and nitrate, other physicochemical parameters of hostel sullage of Federal University of technology, Owerri (FUTO) were above the Environmental Protection Agency (EPA, 2016) and World Health Organisation (WHO, 2011) permissible limits. The high values of the parameters can distort the homeostatic balance of the receiving water. This correlates the work of Adebayo *et al*. (2016) on Ota Mmirri River at FUTO station who suggested that the high values of some physicochemical properties might be from the wastewater discharged from FUTO hostels. The chemical properties of sullage samples from male and female hostels varied within the groups. This might be as a result of the quality of the water used by students, personal habits and lifestyles, the type of cosmetic used and laundry activities in the hostels (Adebayo *et al*., 2016). Onyekuru *et al*. (2017) reported that hand pump wells near septic tank units, which are a major source of water in

FUTO hostels do not impair the quality of water supply in aquifer system as anticipated. The values of the physicochemical parameters of hostel sullage may be traced to the contaminants from shampoos, hair dyes, toothpastes and cleaning chemicals. Since more compounds are chemically oxidized than biologically oxidized (Okereke *et al.*, 2016c; Kiely, 1997), COD values were higher than the BOD values. Sulphate contamination might be from laundry detergents and soaps (Ormiston Association LTD, 2008).

Concentration levels of Heavy Metals in untreated sullage from hostels (chromium, Cr; cadmium, Cd,: lead pb; mercury Hg)

Heavy metal concentration was higher than the EPA and WHO permissible limits and there were no significant difference in the sullage samples at $p<0.05$ for the four parameters. Mercury is a serious environmental issue that is of global concern, particularly in developing countries (Akolo & Adebayo, 2014). The mercury could be from skin-lightening soaps and creams possibly used by students. As reported by Okereke *et al.* (2016), mercury is found in some cosmetics, such as eye make-up, cleaning products, and mascara. Cadmium might have been introduced through body and hair creams as an investigation carried by Ayenimo, Adeeyinwo & Amoo (2010) revealed that some hair cream contains cadmium. Incidentally, it is considered to be carcinogenic to humans (Okereke *et al.*, 2016). The presence of lead in the male and combined sullage might be as a result of dye material used for shirts, shoes, and hair since it is known that lead is very high in dye. Lead, which is a constituent of some lipsticks and leaded eye powders, is readily absorbed through the skin (Okereke, *et al*., 2016). Onyekuru, Nwankwoala & Uzor (2017), reported the presence of these heavy metals in Ota Mmirri River, FUTO Station. This result tends to agree with their suspicion that heavy metals in Ota Mmirri River were introduced from FUTO hostels since there is no industry that uses metals as raw materials for production or as by-products nearby. This is also in agreement with the work of Adebayo, Ebbeniro, Onyediran & Oluwatatosun (2016).

Microbial load of untreated sullage from hostels

The THC and TCC for bacteria in table 4.3 were high. *Micrococcus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp and *Bacillus* sp were present in the sullage, in agreement with a report by WHO (2006). Eze *et al.* (2015), carried out microbiological and physicochemical characteristics of greywater samples in Umuahia, an obtained the total heterotrophic plate count range of 8.1 x 105 ± 0.04 - 1.11 x 106 ± 0.40 cfu/ml and microbal isolates such as *Enterobacter aerogenes, Proteus* species*, Escherichia coli, Pseudomonas aeruginosa, Salmonella* species*, Staphylococcus aureus, Rhizopus* species*, Penicillium* species*, Aspergillus* species *and Trichoderma* species. Results obtained are also in agreement with those of Abdalrahman *et al.*, (2015). The bacterial load is in agreement with Ogah & Ogah, (2018), where they assessed the water from Ota Mmirri River, FUTO Station in Owerri, Imo State. In a related study by Ogah & Ogah (2018), carried out in Ota Mmiri River, FUTO, a total count ranged from $1.2x10^5$ - $9.0x10^5$ cfu/ml and total coliform count range of $5.0x10^5$ and $9.0x10^5$ cfu/ml were obtained. These values might be as a result of self purification as the river flows. Duru *et al.* (2012) also observed high bioload levels of $4.0x10^4$ - $5.1x10^6$ cfu/ml from Ota Mmirri River in addition to *proteus* sp and *vibrio* sp among the bacterial isolates mentioned above. *Saccharomyces* sp & *Penicillium* sp were the only fungi found in the sullage as shown in Table 4.6. Their presence might be as a result of underwear washing, presence of organic carbon, solids and high concentrations of fats, oils and grease. The high microbial counts and the presence of pathogenic microorganism in the sullage showed that there is a need to treat FUTO sullage before discharge into Ota Mmiri River. *Salmonella* sp, *Shigella* sp indicates faecal contamination in water. *Enterobacter* sp (eg *E.coli*) has been implicated as a causative agent of water borne diseases. The presence of *Staphylococcus* sp signifies contamination from human as *Staphylococcus* sp is a normal flora of the human body. These have already been reported by Ogbulie, Ogbulie & Umezuruike (2008).

Chemical parameters of sullage treated with absorbents

From Table 4.6, there was a reduction in the physicochemical properties of the sullage after the treatment with activated carbon forms of the agrowastes. Apart from BOD, their levels were below the EPA and WHO discharge limit, thus does not pose any threat to environment. Egdon *et al.* (2013) reported that chemically activated carbon from maize cob produced at 250^oC reduced the physicochemical parameters like pH, COD, BOD and nitrate in salon wastewater which was attributed to the unique features like large surface area, high degree of surface reactivity and favorable pore size.

Concentration levels of Heavy Metals in sullage treated with adsorbents

In this study, there was a reduction in the heavy metals parameters of the sullage after the treatment with activated carbon from the agrowastes. Mercury levels surpassed the EPA and WHO discharge limits. Lead was slightly above the discharge limit while cadmium and chromium were below the discharge limits. Several works done on chemically activated rice showed it as a promising precursor for treatment of wastewater laden with heavy metals for the removal of lead, mercury, cadmium, and chromium (Chuah, Jumasiah, Azni, Kayaton, & Thomas-Choong, 2015; Mohan & Sreelakshmi, 2008). Egdon (2013), also reported on the use of chemically activated corn cob to remove heavy metals like zinc, chromium, cadmium and iron.

Microbial load of sullage treated with adsorbents

From Table 4.8, the microbial load reduced while *Bacillus* sp after treatment with the adsorbent. This study suggested that the adsorbents have antibacterial activity. Nuhu, Omali & Clifford (2018) reported that agricultural waste-based activated carbon contains antimicrobial activity. In a related work by Carmalin (2013), rice husk and coconut husk was found to be effective in decontaminating water loaded with bacteria. Lelili & Detlef (2005) reported that microorganisms attach to activated carbon through strong LiftShitz and vanderWaals forces.

Efficiency of the adsorbents in the removal of Chemical Properties from sullage

Nitrate was better adsorbed by the activated carbon followed by phosphate and sulphate, then chlorine. The efficiency of removal was in this order: Rice husk + corn cob +coconut husk > rice husk + Corn cob > rice husk > rice husk + coconut husk > corn cob > coconut husk > corn cob+ coconut husk and rice husk + coconut husk. For their individual strengths, activated carbon performed in this order: rice husk > corn cob > coconut husk. In combination, rice husk + corn cob + coconut husk > rice husk + corn cob > corn cob + coconut husk > rice husk + coconut husk. A combination of rice husk, corn cob and coconut husk best adsorbed the four inorganic compounds. Zahid (2016) reported that chemically activated rice husk had high efficiency in removing inorganic compounds in wastewater.

Efficiency of the adsorbents in the removal of heavy metals from sullage

Chromium removal efficiency was 100%. Chromium (Cr) was removed more by the adsorbents, followed by cadmium (Cd), lead (Pb) and mercury (Hg).Cr was 100% removed. Cd percentage removal ranged from 89.47 - 100%, Pb ranged from 0 - 100% while mercury was 6.24 – 40.2%. Mercury had the least percentage removal by the adsorbents. Efficiency removal was in this order: rice husk > rice husk + corn cob > corn cob > coconut husk > corn cob + coconut husk > rice husk + corn cob + coconut husk > rice husk + coconut husk. For their individual strengths, activated carbon performed in this order: rice husk > corn cob > coconut husk. In combination, rice husk + corn cob > corn cob + coconut husk > rice husk + corn cob + coconut husk > rice husk + coconut husk. Buah, Maccarthy & Ndu (2016), reported the efficiency for chemically activated corn cob in removing lead, was 99.6%; cadmium, 99.7%

and copper was 99%. Jabber *et al*. (2016) reported the efficiency of rice for cadmium, lead and chromium from aqueous solutions as 97.96%, 90% and 84% respectively.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATION

5.1 CONCLUSION

There was no significant difference ($p \leq 0.05$) between the chemical, heavy metals and microbial properties of sullage samples from male, female and the combination of both hostels of Federal University of Technology, Owerri. However, there was a significant difference between the untreated sullage (MF) and the treated sullage samples. Some of the chemical parameters (COD, BOD, SO_4^{2-}), and all the heavy metals and microbial parameters investigated were above EPA and WHO discharge limits. NO_3^-, Cl^- and pH were observed to be within the EPA and WHO discharge limits. Total Heterotrophic Count of $6.9 x 10^{10}$ – $1.2 x 10^{11}$ and Total Coliform Count of $3.7 x 10^6$ - $6.4 x 10^6$, with presence of *Bacillus* sp, *Micrococcus* sp, *Staphylococcus* sp, *Enterobacter* sp, *Salmonella* sp, *Shigella* sp revealed that the sullage generated were loaded with pathogenic bacteria. Therefore, FUTO hostel sullage is not safe to be discharged into the environment. High removal efficiencies observed in the chemical and heavy metal parameters; complete elimination of *Bacillus* sp and significant reduction of the microbial load revealed that activated carbons from rice husk, corn cob and coconut husk can be used singly or in combined states for the treatment of sullage.

5.2 RECOMMENDATION

Agrowastes can be used in treatment of wastewater particularly the activated carbon forms. Work done in laboratory conditions could be extended to the field with the intention of assessing the applicability.

Combined efforts of stakeholders in the environment particularly sanitary engineers should harness the results obtained to see how agrowastes can be used to construct inbuilt treatment devices in homes in order to achieve some degree of treatment in household wastewater before discharge.

The consortium efficiency of other activated carbon forms of agrowastes should be investigated.

Factors which may affect the efficiency of the agrowastes in effluent treatment should be critically studied.

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APPENDICES

Appendix 1

Heavy metals, chemical and microbial characterization of sullage before treatment.

Before Treatment			
Element	M	F	MF
Hg(ppm)	2.65	4.01	2.68
Cr	0.14	-2.51	0.13
Cd	0.071	0.072	0.076
Pb	0.202	-0.02	0.06
COD	486.2	342.2	395.5
BOD	175.03	123.2	142.37
PH	7.33	7.01	7.27
PO4	7.68	4.25	7.26
Cl	31	24.85	21.89
NO3	28.63	7.74	13.83
SO4	347.08	319.58	289.51
T H C	6.90E+10	1.20E+11	8.90E+10
T C C	4.90E+06	6.40E+06	1.50E+06
T F C	1.50E+08	2.20E+10	1.60E+10

Appendix 2

Heavy metals, chemical and microbial characterization of sullage after treatment.

After treatment

Element	rice	corncob	Coconuthusk	rice + corncob	rice + coconut husk	corncob + coconuthusk	rice + corncob + coconuthusk
Hg(ppm)	1.73	2.51	2.18	2.24	2.27	1.6	2.16
Cr	0	0	0	0	0	0	0
Cd	0	0	0	0.001	0	0	0.008
Pb	0	0	0.06	0	0.06	0.03	0.02
COD	158.7	190.5	188.1	170.1	127.8	187.7	122.4
BOD	57.1	68.6	67.7	61.3	46	67.6	44
PH	7.13	7.17	7.17	7.11	7.106	7.11	7.07
PO4	3.61	5.6	5.34	4.89	6.63	6.6	3.89
Cl	20.47	17.28	20.24	20.94	18.46	17.75	21.13
NO3	8.8	12.49	4.11	0	12.79	11.13	0
SO4	147.8	175.4	251.7	178.7	163.2	183.9	117.7
T H C	5.30E+10	7.30E+09	7.60E+10	1.90E+10	3.90E+10	3.98E+10	1.60E+09
T C C	2.30E+06	7.30E+08	1.10E+06	3.60E+05	3.90E+05	2.20E+05	6.50E+05
T F C	1.67E+08	7.30E+08	1.30E+08	2.60E+08	1.00E+08	1.20E+09	2.00E+08

Appendix 3

ANOVA Results for Chemical Analysis

Univariate Analysis of Variance

Untreated

[DataSet3] C:\Users\CHIDIMMA\Documents\Chemical analysis.sav

Between-Subjects Factors

		Value Label	N
Chemical	1	COD	3
	2	BOD	3
	3	P4	3
	4	PO4	3
	5	CL	3
	6	NO3	3
	7	SO4	3
Sullage	1	M	7
	2	F	7
	3	MF	7

Tests of Between-Subjects Effects

Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	370514.674	1	370514.674	142.617	.007
	Error	5195.939	2	2597.969[a]		
Chemical	Hypothesis	501391.805	6	83565.301	115.024	.000
	Error	8718.036	12	726.503[b]		
Sullage	Hypothesis	5195.939	2	2597.969	3.576	.061
	Error	8718.036	12	726.503[b]		
Chemical * Sullage	Hypothesis	8718.036	12	726.503	.	.
	Error	.000	0	.[c]		

a. MS(Sullage)
b. MS(Chemical * Sullage)
c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Sullage)	Var(Chemical * Sullage)	Var(Error)	Quadratic Term
Intercept	7.000	1.000	1.000	Intercept, Chemical
Chemical	.000	1.000	1.000	Chemical
Sullage	7.000	1.000	1.000	
Chemical * Sullage	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.
b. Expected Mean Squares are based on the Type III Sums of Squares.

Treated

Univariate Analysis of Variance

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Between-Subjects Factors

		Value Label	N
Chemical	1	COD	7
	2	BOD	7
	3	PH	7
	4	PO4	7
	5	CL	7
	6	NO3	7
	7	SO4	7
Treatment	2	Rh	7
	3	Cc	7
	4	Ch	7
	5	Rh+Cc	7
	6	Rh+Ch	7
	7	Cc+Ch	7
	8	Rh+Cc+Ch	7

Tests of Between-Subjects Effects

Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	189600.031	1	189600.031	234.963	.000
	Error	4841.607	6	806.935[a]		
Chemical	Hypothesis	237689.564	6	39614.927	128.490	.000
	Error	11099.250	36	308.313[b]		
Treatment	Hypothesis	4841.607	6	806.935	2.617	.033
	Error	11099.250	36	308.313[b]		
Chemical * Treatment	Hypothesis	11099.250	36	308.313	.	.
	Error	.000	0	.[c]		

a. MS(Treatment)
b. MS(Chemical * Treatment)
c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Treatment)	Var(Chemical * Treatment)	Var(Error)	Quadratic Term
Intercept	7.000	1.000	1.000	Intercept, Chemical
Chemical	.000	1.000	1.000	Chemical
Treatment	7.000	1.000	1.000	
Chemical * Treatment	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.
b. Expected Mean Squares are based on the Type III Sums of Squares.

Appendix 4

T-Test for chemical analysis

[DataSet1] C:\Users\CHIDIMMA\Documents\chemical analysis.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Rh	7	57.65857	67.798345	25.625366

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	5.577	.036	1.038	12	.320	67.7171	65.2518	-74.4544	209.888691
	Equal variances not assumed			1.038	8.118	.329	67.7171	65.2518	-82.37478	217.809079

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Cc	7	68.14857	81.434169	30.779223

Independent Samples Test

	Levene's Test for Equality of Variances		t-test for Equality of Means					
	F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference

									Lower	Upper
Mean Values	Equal variances assumed	3.775	.076	.849	12	.413	57.227143	67.442597	-89.717653	204.171938
	Equal variances not assumed			.849	8.953	.418	57.227143	67.442597	-95.462015	209.916300

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\ chemical analysis.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Ch	7	77.76571	101.235468	38.263410

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	1.904	.193	.669	12	.516	47.6100	71.170443	-107.458	202.677074
	Equal variances not assumed			.669	10.187	.518	47.6100	71.170443	-110.575	205.794529

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\ chemical analysis.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean

Mean_Values	MF	7	125.37571	158.770285	60.009527
	Rh+Cc	7	63.29143	78.631740	29.720004

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean Values	Equal variances assumed	4.109	.065	.927	12	.372	62.084	66.9658	-83.8217	207.9903
	Equal variances not assumed			.927	8.776	.379	62.084	66.9658	-89.9936	214.1621

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Rh+Ch	7	54.56943	64.339115	24.317900

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	T	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_ Values	Equal variances assumed	6.095	.030	1.094	12	.296	70.806	64.7495	-70.2709	211.8834

	Equal variances not assumed			1.094	7.919	.306	70.8062	64.7495	-78.7733	220.3858

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Cc+Ch	7	68.82714	82.657004	31.241411

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean Values	Equal variances assumed	3.630	.081	.836	12	.420	56.5486	67.654	-90.858529	203.955672
	Equal variances not assumed			.836	9.030	.425	56.5486	67.654	-96.420161	209.517304

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	7	125.37571	158.770285	60.009527
	Rh+Cc+Ch	7	45.17000	53.229600	20.118898

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	7.976	.015	1.267	12	.229	80.2057	63.292285	-57.696	218.108
	Equal variances not assumed			1.267	7.332	.244	80.2057	63.292285	-68.094	228.505

Appendix 5
ANOVA Results for Heavy metals

Univariate Analysis of Variance

Untreated

[DataSet2] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis.sav

Between-Subjects Factors

		Value Label	N
Heavy_Metals	1	Hg	3
	2	Cr	3
	3	Cd	3
	4	Pb	3
Sullage	1	M	4
	2	F	4
	3	MF	4

Tests of Between-Subjects Effects
Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	4.764	1	4.764	26.966	.035
	Error	.353	2	.177[a]		
Heavy_Metals	Hypothesis	26.023	3	8.674	9.390	.011
	Error	5.542	6	.924[b]		
Sullage	Hypothesis	.353	2	.177	.191	.831
	Error	5.542	6	.924[b]		
Heavy_Metals * Sullage	Hypothesis	5.542	6	.924	.	.
	Error	.000	0	.[c]		

a. MS(Sullage)
b. MS(Heavy_Metals * Sullage)
c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Sullage)	Var(Heavy_Metals * Sullage)	Var(Error)	Quadratic Term
Intercept	4.000	1.000	1.000	Intercept, Heavy_Metals
Heavy_Metals	.000	1.000	1.000	Heavy_Metals
Sullage	4.000	1.000	1.000	
Heavy_Metals * Sullage	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.
b. Expected Mean Squares are based on the Type III Sums of Squares.

Treated

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Between-Subjects Factors

		Value Label	N
Heavy_Metals	1	Hg	7
	2	Cr	7
	3	Cd	7
	4	Pb	7
Treatment	2	Rh	4
	3	Cc	4
	4	Ch	4
	5	Rh+Cc	4
	6	Rh+Ch	4
	7	Cc+Ch	4
	8	Rh+Cc+Ch	4

Tests of Between-Subjects Effects

Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	7.896	1	7.896	304.695	.000
	Error	.155	6	.026[a]		
Heavy_Metals	Hypothesis	22.936	3	7.645	297.599	.000
	Error	.462	18	.026[b]		
Treatment	Hypothesis	.155	6	.026	1.009	.450
	Error	.462	18	.026[b]		
Heavy_Metals * Treatment	Hypothesis	.462	18	.026	.	.
	Error	.000	0	.[c]		

a. MS(Treatment)
b. MS(Heavy_Metals * Treatment)

c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Treatment)	Var(Heavy_Metals * Treatment)	Var(Error)	Quadratic Term
Intercept	4.000	1.000	1.000	Intercept, Heavy_Metals
Heavy_Metals	.000	1.000	1.000	Heavy_Metals
Treatment	4.000	1.000	1.000	
Heavy_Metals * Treatment	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.
b. Expected Mean Squares are based on the Type III Sums of Squares.

Appendix 6
T-Test for heavy metals analysis

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Rh	4	.43250	.865000	.432500

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	Df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	.687	.439	.390	6	.710	.304000	.779082	-1.602345	2.210345
	Equal variances not assumed			.390	5.230	.712	.304000	.779082	-1.672467	2.280467

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Cc	4	.62750	1.255000	.627500

Independent Samples Test

	Levene's Test for Equality of Variances		t-test for Equality of Means					
	F	Sig.	t	Df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference

									Lower	Upper
Mean_ Values	Equal variances assumed	.005	.948	.121	6	.908	.109000	.902036	-2.0982	2.3162
	Equal variances not assumed			.121	5.994	.908	.109000	.902036	-2.0988	2.3168

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Ch	4	.56000	1.080370	.540185

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	Df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	.147	.715	.209	6	.841	.176500	.843630	-1.887789	2.240789
	Equal variances not assumed			.209	5.812	.841	.176500	.843630	-1.904112	2.257112

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Rh+Cc	4	.56025	1.119833	.559917

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	Df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	.095	.769	.206	6	.844	.176250	.856399	-1.919282	2.271782
	Equal variances not assumed			.206	5.876	.844	.176250	.856399	-1.930021	2.282521

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Rh+Ch	4	.58250	1.125355	.562678

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_ Values	Equal variances assumed	.089	.776	.179	6	.863	.154000	.858207	-1.945956	2.253956
	Equal variances not assumed			.179	5.884	.864	.154000	.858207	-1.956011	2.264011

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Cc+Ch	4	.40750	.795126	.397563

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_ Values	Equal variances assumed	.974	.362	.433	6	.680	.329000	.760242	-1.531246	2.189246

	Equal variances not assumed			.433	4.978	.683	.329000	.760242	-1.627847	2.285847

T-Test

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	4	.73650	1.296013	.648006
	Rh+Cc+Ch	4	.54700	1.075365	.537682

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	T	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean Values	Equal variances assumed	.154	.708	.225	6	.829	.189500	.842030	-1.8709	2.249
	Equal variances not assumed			.225	5.802	.830	.189500	.842030	-1.888	2.266

Appendix 7
ANOVA Results for Microbial Analysis

Univariate Analysis of Variance

Untreated

[DataSet4] C:\Users\CHIDIMMA\Documents\Microbia analysis.sav

Between-Subjects Factors

		Value Label	N
Microbial	1	THC	3
	2	TCC	3
	3	TFC	3
Sullage	1	M	3
	2	F	3
	3	MF	3

Tests of Between-Subjects Effects
Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	11106546233759985000000.000	1	11106546233759985000000.000	25.110	.038
	Error	884631378646666700000.000	2	442315689323333350000.000[a]		
Microbial	Hypothesis	15139927987519985000000.000	2	7569963993759993000000.000	43.826	.002
	Error	690916967293333300000.000	4	172729241823333320000.000[b]		
Sullage	Hypothesis	884631378646666700000.000	2	442315689323333350000.000	2.561	.192
	Error	690916967293333300000.000	4	172729241823333320000.000[b]		
Microbial * Sullage	Hypothesis	690916967293333300000.000	4	172729241823333320000.000	.	.
	Error	.000	0	.[c]		

a. MS(Sullage)
b. MS(Microbial * Sullage)
c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Sullage)	Var(Microbial * Sullage)	Var(Error)	Quadratic Term
Intercept	3.000	1.000	1.000	Intercept, Microbial
Microbial	.000	1.000	1.000	Microbial
Sullage	3.000	1.000	1.000	
Microbial * Sullage	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.
b. Expected Mean Squares are based on the Type III Sums of Squares.

Univariate Analysis of Variance

Treated

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Between-Subjects Factors

		Value Label	N
Microbial	1	THC	7
	2	TCC	7
	3	TFC	7
Treatment	2	Rh	3
	3	Cc	3
	4	Ch	3
	5	Rh+Cc	3
	6	Rh+Ch	3
	7	Cc+Ch	3
	8	Rh+Cc+Ch	3

Tests of Between-Subjects Effects

Dependent Variable: Mean_Values

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	2725103564422885400000.000	1	2725103564422885400000.000	11.939	.014
	Error	1369469367461325300000.000	6	228244894576887560000.000[a]		
Microbial	Hypothesis	5212438953205744000000.000	2	2606219476602872000000.000	11.159	.002
	Error	2802546311162649000000.000	12	233545525930220750000.000[b]		
Treatment	Hypothesis	1369469367461325300000.000	6	228244894576887560000.000	.977	.481
	Error	2802546311162649000000.000	12	233545525930220750000.000[b]		
Microbial * Treatment	Hypothesis	2802546311162649000000.000	12	233545525930220750000.000	.	.
	Error	.000	0	.[c]		

a. MS(Treatment)

b. MS(Microbial * Treatment)

c. MS(Error)

Expected Mean Squares[a,b]

Source	Variance Component			
	Var(Treatment)	Var(Microbial * Treatment)	Var(Error)	Quadratic Term
Intercept	3.000	1.000	1.000	Intercept, Microbial
Microbial	.000	1.000	1.000	Microbial
Treatment	3.000	1.000	1.000	
Microbial * Treatment	.000	1.000	1.000	
Error	.000	.000	1.000	

a. For each source, the expected mean square equals the sum of the coefficients in the cells times the variance components, plus a quadratic term involving effects in the Quadratic Term cell.

b. Expected Mean Squares are based on the Type III Sums of Squares.

Appendix 8

T-Test for microbial analysis

[DataSet1] C:\Users\CHIDIMMA\Documents\Heavy Metals analysis2.sav

Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Rh	3	17723100000.00000	30550802554.597480	17638514078.855960

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	1.138	.346	.530	4	.624	17277400000.000000	32579641073.733967	-73178184988.269640	107732984988.269640
	Equal variances not assumed			.530	3.415	.628	17277400000.000000	32579641073.733967	-79625953954.076390	114180753954.076390

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Cc	3	2920000000.00000	3793191268.575841	2190000000.000000

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	10.638	.031	1.167	4	.308	32080500000.000000	27479300092.675820	-44214268236.571040	108375268236.571040
	Equal variances not assumed			1.167	2.026	.362	32080500000.000000	27479300092.675820	-84734695421.185620	148895695421.185600

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Ch	3	25377033333.33333	43840822521.975260	25311510684.557026

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	.029	.873	.258	4	.809	9623466666.666666	37295956975.492370	-93926710521.606610	113173643854.939940
	Equal variances not assumed			.258	3.975	.809	9623466666.666666	37295956975.492360	-94180941081.066930	113427874414.400250

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Rh+Cc	3	6420120000.00000	10895269103.753242	6290386549.945348

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean Values	Equal variances assumed	7.144	.056	1.017	4	.367	28580380000.000000	28104889192.648790	-49451302031.056080	106612062031.056080
	Equal variances not assumed			1.017	2.210	.407	28580380000.000000	28104889192.648790	-81959387615.269130	139120147615.269130

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Rh+Ch	3	13033463333.33333	22487735554.831512	12983300176.047092

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	2.885	.165	.725	4	.509	21967036666.666668	30313065121.242320	-62195524612.748520	106129597946.081850
	Equal variances not assumed			.725	2.855	.523	21967036666.666668	30313065121.242320	-77324817774.544140	121258891107.877490

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Cc+Ch	3	13666740000.00000	22640016048.642723	13071219360.141323

Independent Samples Test

	Levene's Test for Equality of Variances	t-test for Equality of Means

		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	2.842	.167	.703	4	.521	21333760000.000000	30350825510.095547	-62933640926.217090	105601160926.217090
	Equal variances not assumed			.703	2.866	.535	21333760000.000000	30350825510.095547	-77862926251.553020	120530446251.553020

T-Test

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Group Statistics

	Treatment	N	Mean	Std. Deviation	Std. Error Mean
Mean_Values	MF	3	35000500000.00000	47444151386.129776	27391893574.255383
	Rh+Cc+Ch	3	600216666.66667	871556160.458598	503193183.854648

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mean_Values	Equal variances assumed	12.197	.025	1.256	4	.278	34400283333.333	27396515051.44	-41664636780.71	110465203447.37
	Equal variances not assumed			1.256	2.001	.336	34400283333.33	27396515051.44	-83401250831.59	152201817498.26