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Comparative Assessment of Disinfection Efficiency of $ZnCl_2$-activated Coconut Shell and Sodium Hypochlorite on River Water Sample

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Abstract

This study was carried out to compare the disinfection efficiencies of granulated ZnCl2-activated coconut shell carbon (GACSC) and sodium hypochlorite (NaOCl) on a River water sample. The coconut shell was carbonized at a temperature range of 450 – 650^{o}C and thereafter was activated with 0.1 M ZnCl2 solution at different impregnation ratios. Characterization of the (GACSC) was carried out to assess the morphology, elemental composition as well as pore development using a scanning electron microscope, energy dispersive X-ray, and iodine value test, respectively. A 250 ml of the water sample was separately disinfected in conical flasks with equal masses of both GACSC and NaOCl. Results obtained showed an increase in the pore volume and pore distribution after activation, while the percentage composition of oxygen decreased and that of carbon increased. The optimum value obtained for the iodine value test was 560 mg/g at a 1:2 impregnation ratio. The values obtained after the disinfection indicated that both the GACSC and NaOCl were able to remove microbes from the untreated water samples. However, 0.4 g of GACSC gave a higher disinfection efficiency of 90 % than 0.4 g of NaOCl which gave 30 % efficiency of removal. Bacillus spp. was removed from the samples by both GACSC and NaOCl but was not able to remove completely all the Escherichia coli in the water samples. The results revealed that GACSC demonstrated more potential for water disinfection than NaOCl. Therefore, it is suggested that GACSC be used as an alternative to NaOCl to disinfect drinking water.

Keywords: Microorganism, granulated activated coconut shell, sodium hypochlorite, disinfection efficiency, byproduct of disinfection

1. Introduction

Water is only potable when it is free of contaminants such as microorganisms, dissolved organic compounds, inorganic substances as well as suspended materials. Water bodies (both surface water and groundwater) have remained a sink for volumes of wastes generated by domestic, industrial, municipal, and mining activities. In developing countries where there is lack of strict enforcement of policies and political willpower by the governments, generated wastes are indiscriminately discharged into the environment without pretreatment to remove contaminants. The consequence of this has been a continuous dearth of potable water resulting in several waterborne diseases such as cholera, typhoid, dysentery, and hepatitis A. Furthermore, most of the urban cities and urban-poor lack proper planning, in that there are few or no facilities for evacuation of solid wastes from refuse dumpsites and effluents from homes.

There has been a global concern for the provision of potable water as a result of the increasing human population (Izah *et al.,* 2016). This has led to the development of techniques such as reverse osmosis, dialysis, membrane filtration, ozonolysis, electrocoagulation, UV-radiation, and chlorination to treat water. Conventionally, the use of chlorine as a chemical agent to disinfect water has found a wide application due to its ability to kill or inactivate a wide *variety of microbial pathogens including Escherichia coli, Pseudomonas spp., Klebsiella spp.,* Bacillus spp., *Staphylococcus aureus*, and others. The ineffectiveness of the use of chlorination methods to disinfect some viruses and bacteria has been studied (Mohammed, 2019). Sodium hypochlorite and other oxidative and non-oxidative chlorination react with organic matter contained in the source water to form a by-product of disinfection (BPDs) (Kamel *et al.,* 2009; Nsikak *et al.,* 2017).

Avalanche of studies have shown that granular activated carbons, due to their high surface area, surface morphology and chemistry, and elemental compositions, pick up contaminants easily in water by adsorption process and do not form by-products (Rivera-Utrilla *et al*., 2001; Tong *et al*., 2010; Ekta *et al*., 2012; Mohammed *et al*., 2016). Furthermore, they generate low waste, have a low cost of production, and are renewable; hence adsorption of pathogenic microorganisms from water samples using activated carbon has been applied by several researchers (Ekta *et al.,* 2012; Mohammed *et al*., 2016; Ding *et al*., 2020; King *et al.,* 2020; Dana *et al.,* 2021; David *et al*., 2021a). Quite a lot of studies have been carried out on both the adsorption and disinfection of pathogenic bacteria in water samples with granulated activated carbon by filtration technique and with sodium hypochlorite, respectively, but there are dearth of works in literature that compared the disinfection capabilities of activated carbon and sodium hypochlorite in water samples. Therefore, this research aimed to comparatively assess the disinfection efficiencies of $ZnCl_2$-activated coconut shells and Sodium hypochlorite in River water.

2.1 Materials and Methods

2.1.1 Preparation of Sample Bottles and Sampling

One liter (1 L) plastic bottle was filled with liquid soap and allowed to soak for 24 hours before thoroughly washing the caps and bottles and rinsing them severally with distilled water. There were autoclaved at 121^{o}C using an autoclave sterilizer (Techmel and Techmel, USA) for

15 minutes, then capped and wrapped with foil to avoid contamination. The preparation of sample bottles and sampling of the River water followed standard guidelines. River water samples were collected at Nwakpu River, Ndufu –Alike, Ikwo. The River lies at Longitude 8o17′0″E and Latitude 6o8′0″N (David *et al*., 2021b). The water was collected at the two banks and in the middle of the River using the already sterilized 1-liter containers. The sample bottles were rinsed three times with the River sample and dipped 30 centimeters below the water surface. They were collected against the water current and flow to avoid air bubbles (David *et al.,* 2021). Composite samples were pooled from the three sampling points into a 20 L container autoclaved and some parameters like the temperature and pH were measured in situ. The composite sample was then tightly covered with the sterilized cap and foil to avoid contamination. It was then transported to the laboratory in an ice bag and stored in a refrigerator at 4^{o}C before disinfection.

2.1.2 Preparation of Activated Carbon

The coconut shell was packed in large quantities from a local market, Iziogo Inyimegu, in Ebonyi State. The preparation of the activated carbon was done according to the procedure and method described by David *et al*. (2021a).

2.1.3 Activation/Development of Pores

The activation was carried out by adapting the methods of David et al. (2021a). Different masses of the carbonized coconut shell were soaked for 24 hours with 0.1M ZnCl2 solution to give impregnation ratios of 1:1, 1:2, 1:3, and 2:1. They were filtered and dried in an oven for 2 hours after which the activated biomaterial was washed thoroughly with distilled water and dried again in the oven (Gen lab) for another 2 hours.

2.2 Characterization of activated Carbon

2.2.1 Water Physicochemical Parameters

The pH and temperatures of the River samples were measured in situ with Orion Star A221 pH portable meter and Platinum Ultra-Accurate Digital Thermometer.

2.2.2 Iodine Value Test

The process adopted the technique of David et al. (2021a). A 1.0 g of the different impregnated ratios of the coconut shell carbon was separately mixed with 5 ml of 5 % w/v of concentrated hydrochloric acid in a 100 ml conical flask and mechanically shaken to wet the adsorbent. This was then followed by adding 25 ml of prepared 0.1 M iodine solution and the mixture was stirred for 1 hour using temperature controlled electric magnetic stirrer. The 25 ml filtrate was titrated against a standardized 0.1 M sodium thiosulphate solution until the filtrate turns cloudy using starch solution as an indicator.

$$\text{Iodine value} = \left[(\text{Mi x } 126.93 \text{ x Vi}) - \left(\frac{V_i+V_a}{VF}\right)\left(\frac{M_t+126.93}{W_{ac}}\right)\right] \qquad \text{-------- (1)}$$

The iodine values of the different impregnated biomaterials were obtained using equation (1)

Where: Mi = molar concentration of iodine solution, Vi = Volume of iodine solution, Va = Volume of 5 % HCl used, VF = Volume of filterate used, Vt = Volume of average titre, Mt = Molar concentration of thiosulfate used, Wac = Weight of adsorbent

2.2.3 Scanning Electron Microscope (SEM)

Scanning electron microscope (Phenom-Prox, Phenom world Eindhoven the Netherlands) was used for the surface and morphological characterization.

2.2.4 Energy Dispersive X-ray (EDX)

EDX is an analytical technique used for the elemental analysis or chemical characterization of a sample. It relies on the investigation of the interaction of some source of X-ray excitation and a sample.

2.3 Disinfection of the River Water Samples

The same doses (0.2, 0.4, 0.6, 0.8, and 1 g) of NaOCl and the $ZnCl_2$ -activated coconut shell carbon was used in 250 ml of the river water sample and disinfected for 1 hour at an ambient condition keeping the solution stirring in a magnetic stirrer.

2.4 Culturing of Treated Water Samples for Microbial Analysis/Enumeration and Isolation of Bacteria from Water Sample

The enumeration of bacteria present in the water sample was carried out by transferring 0.1 ml of water sample into a sterile plate containing Nutrient Agar and Eosine Methylene blue agar already, sterilized in the autoclave at 121 ^{o}C for 15 minutes in the incubator (mereck, Germany) and allowed to cool to around 55 ^{o}C. A sterile spreader was used to spread the 0.1 ml of water on the entire surface of the nutrient Agar. Plates were then incubated at 37 ^{o}C for 24 hours. The colonies which developed were counted using the colony counter (mereck, Germany).Discrete colonies from the culture plates were randomly picked using a sterile inoculating wire loop and sub-cultured for purification by streaking on nutrient Agar plates. The culture was incubated at 37 ^{o}C for 24 hours (Aleruchi *et al*., 2022).

A culture of isolates on nutrient broth was subcultured on Eosine Methylene blue Agar, Mannitol salt Agar (MSA), and cetrimide Agar. These selective media were employed for selective growth

and differentiation of isolate for the purpose of identification. Mannitol salt Agar (MSA) was used for the selective isolation of enteric Staphylococcus aerus, while Centrimide Agar was used for *E. coli* differentiation. The cultures were incubated at 40 ^{o}C and 37 ^{o}C, respectively for 24 hours. Confirmation of bacteria isolates was based on their phenotypic characteristics and standard cultural characteristics.

3. Results and Discussion

3.1 Characterization of Granulated Activated Coconut Shell

Table 1 presents the pHs and the temperatures of the River water measured in-situ. It indicated that the River water was slightly acidic. The pH and temperature values obtained fell within WHO permissible limits between 6.5 and 8.5, and 20 and 31.6, respectively, for drinking water.

3.1.2 Effect of Pore Development/Iodine Test Table 2 presents the values of the iodine test carried out to determine the extent of pore development on the surface of the activated coconut shell carbon.

Table 1: pH and Temperature of the River Samples

Sampling Point	pH	Temperature
River Bank 1	6.80	29.00
Middle of River	6.80	30.50
River Bank 2	6.80	29.00
Composite	6.98	29.97

Table 2: Iodine Value Test

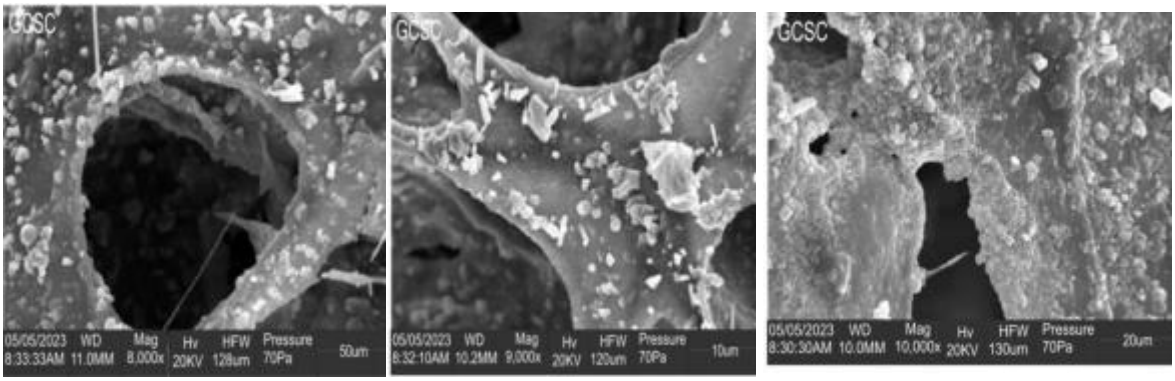
Impregnation Ratio	Iodine Value
1:1	130
1:2	560
1:3	210
2:1	170

At low concentration of $ZnCl_2$ (impregnation ratio of 1:1), there was an insufficient amount of the activation agent to dehydrate the more than enough surface of the carbon material available which led to lower development of pore volume (Molina-Sabio and Rodr´ıguez-Reinoso, 2004). At higher concentrations (impregnation ratio of 1:2) there was a maximum amount of the $ZnCl_2$ to be distributed uniformly on the surface and equally dispersed in the interior of the carbon material to form micropores uniformly. At a much higher concentration (impregnation ratio of 1:3), the

surface of the carbon material available was smaller than the activation agent and hence there was poor development of the pore structures resulting in lower pore volume, hence, a reduction in iodine value. Therefore, the optimum iodine value of 0.056 mg/g was obtained at the impregnation ratio of 1:2. This observation was in line with other investigations (Abdul *et al.*, 2001).

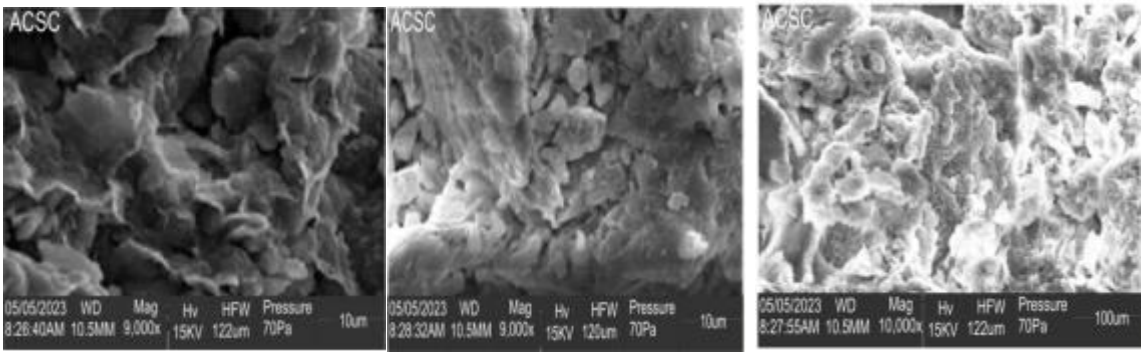
3.1.3 Physical Surface Structures of the Granulated Non-activated and Activated Coconut Shell Carbon

The results of the morphologies of the granulated non-activated and activated coconut shell carbon were shown in plates 1 a, b, c, d, e, and f.



a b c

Plate 1 (a,b, and c): Scanning Electron Micrographs of the Granulated non-activated coconut shell



d e f

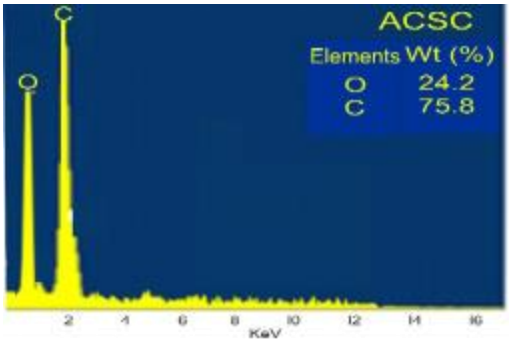
Plate 1 (d, e, and f): Scanning Electron Micrographs of the Granulated Activated Coconut Shell

The morphologies and pore structures of the surfaces of the non-activated and activated coconut shell carbon presented in Plates a, b, c, d, e, and f were investigated using scanning electron micrographs at 8 000, 9 000, and 10 000 magnifications. The results showed the

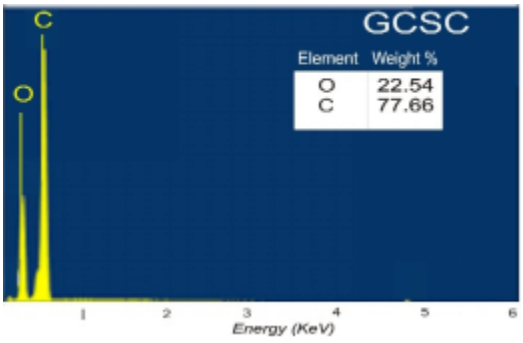
development of cavities and channels after carbonization as a result of the decomposition and escape of volatile components of the lignocellulosic material (Plates a, b, c). The surface of the granulated activated coconut shell at 9,000, and 10,000 magnifications as shown in Plates d, e, and f, were flaky and honeycomb-like with increased pore distribution. The increased pore network and volumes were as a result of the dehydrating action of $ZnCl_2$ and escaping $ZnCl_2$ during the activation process (Cazetta *et al.*, 2011; Yusufu *et al*., 2012; Hesas *et al*., 2013; Iqbaldin *et al.*, 2013; Zhigao *et al*., 2016). This implies that the $ZnCl_2$ activating agent was effective. Similar results were noticed by Hongxia *et al*. (2022).

3.1.4 Energy Dispersive X-ray

The elemental analysis with percentage composition in both the GCSC and GACSC is shown by the energy dispersive X-ray (EDX) in Figure 1(a and b).



(a)



(b)

Figure 1 (a): GCSC-EDX image (b): AGCSC-EDX image The results in Figures 1 (a and b) show the EDX for GCSC and GACSC. From the EDX, the

percentage composition of carbon and oxygen present in the non-activated carbon was 75.8 % carbon and 24.2 % oxygen. After activation, the percentage of oxygen reduced to 22.54 while carbon increased to 77.66 %. This might be attributed to the removal of volatile components of the precursor biomaterial. This depicts that the activation of the coconut shell carbon with $ZnCl_2$ solution was effective due to dehydrating nature of $ZnCl_2$ which removed oxygen atoms previously present in the precursor coconut shell carbon (Malik, 2006; Hock and Zaini, 2018). The implication is that the activated carbon was suitable for adsorption in aqueous solution (Allwar, 2012).

3.2 Microbial Test Result

The results obtained from the microbial analysis for the disinfection of the river water sample using GACSC and NaOCl were shown in Table 3. The result showed the presence of Escherichia coli (*E. coli*), and Bacillus spp. were identified in the non-treated River water sample when a differential selective media, Eosine methylene blue (EMB) was used to culture the water. The presence of *E. coli* in water depicts fecal contamination which shows that the water is not fit for human consumption and for drinking (Chian, 2002; Ekhaise and Omoigberale, 2011).

Table 3: Bacteria Present in the River water samples

Dose (g)/Treatment material	Bacteria Present in the Water
GACSC	
0.2	*Escherichia coli, Pseudomonas* spp.*, and Bacillus* spp.
0.4	*Escherichia coli*
0.6	*Escherichia coli, Bacillus* spp. and *Staphylococcus aureus*
0.8	*Escherichia coli, Pseudomonas* spp.*,* and *Bacillus* spp.
1.0	*Escherichia coli, Pseudomonas* spp.*, and Bacillus* spp.
NaOCl	
0.2	*Escherichia coli,* and *Pseudomonas* spp.
0.4	*Escherichia coli*
0.6	*Escherichia coli, Pseudomonas* spp.*, and Bacillus* spp.
0.8	*Escherichia coli, Pseudomonas* spp.*, and Bacillus* spp.
1.0	*Escherichia coli, Pseudomonas* spp.*, Klebsiella* spp.*, Bacillus* spp. and *Staphylococcus aureus*

After the separate disinfection of the water samples with five separate masses of GACSC and NaOCl, it was observed that more pathogenic bacteria (*Pseudomonas* spp., *Klebsiella* spp., and Staphylococcus aureus) which were not abinitio identified in the non-treated water sample were isolated in the water samples. The contamination of these treated water samples may be implicated in improper handling of the GACSC and NaOCl, as well as exposure to the treated water for a long time before microbial analysis (Theresa *et al.*, 2016). The results indicated that 0.2 g of GACSC did not remove both the *E. coli* and Bacillus *spp*., while with 0.4 g, the reduction

efficiency reached its plateau as there was the total removal of *Bacillus* spp. and achieved 90 % (total coliform count), 28.6 % (total bacteria count) reduction of *E. coli*. With 0.6, 0.8, and 0.1 g of GACSC, it was observed that the reduction efficiency started to decline and reached its minimum with 1.0 g of GACSC. This observation obtained with 0.2 g GACSC has been attributed to less surface area of the GACSC available, while the decline in the adsorption efficiency (both the *E. coli* and *Bacillus* spp. were recalcitrant) after 0.4 g was due to the fact that the micropores available on the surface area of the GACSC have been occupied. Therefore, the adsorption of microorganisms was observed to be dependent on surface area. This is in line with other investigations of other researchers (Saifuddin and Kumaran, 2005; Agbozu and Emoruwa, 2014; Shidvash *et al.*, 2014). On the other hand, the same trend of results was observed when NaOCl was used. With NaOCl, the reduction in the total coliform reached optimum with 0.4 g of the disinfectant, steeped with 0.6 g, increased though insignificantly with 0.8 g, and once again reduced with 1.0 g. Akinbomi and Ikhide (2022) evaluated and compared the disinfectant efficacy of NaOCl and Aloe vera on eight isolated microorganisms from three water samples using the disc diffusion method. The results they obtained indicated that aloe vera and NaOCl were effective as disinfectants against coliform isolated from polluted water sources. However, aloe vera showed more potential to disinfect than NaOCl. From the present study, therefore, 0.4 g of both the GACSC and NaOCl were taken as the optimum dose. However, in Table 4, it can be inferred that GACSC disinfected the River water sample more efficiently than NaOCl even though the total coliform count of 0.1x101 (CFU/ml) was above WHO permissible limits of 0.0 (CFU/ml) (David *et al*., 2021 b). Several workers have shown that activated carbon does not totally remove pathogenic bacteria below the WHO allowable limit from drinking water (David *et al*., 2021 b).

Table 4: Reduction Efficiency in Total Bacterial Counts

Dose (g)/ Treatment Material	**Treated River Sample**	**Untreated River Sample**	**Reduction Efficiency (%)**	**Treated River Sample**	**Untreated River Sample**	**Reduction Efficiency (%)**
	Total coliform count CFU/ml			**Total coliform count CFU/ml**		
GACSC						
0.2	6.0×10^1	1.0×10^1	- 500	1.84×10^3	1.4×10^3	- 31.4
0.4	0.1×10^1	1.0×10^1	90	1.0×10^3	1.4×10^3	28.6
0.6	1.0×10^2	1.0×10^1	- 900	1.3×10^3	1.4×10^3	7.1
0.8	1.4×10^2	1.0×10^1	- 1,300	3.0×10^3	1.4×10^3	- 114.3
1.0	2.0×10^1	1.0×10^1	- 100	4.0×10^3	1.4×10^3	- 185.7
NaOCl						
0.2	0.9×10^1	1.0×10^1	10	1.4×10^3	1.4×10^3	0.0
0.4	0.7×10^1	1.0×10^1	30	1.2×10^3	1.4×10^3	14.3
0.6	1.7×10^2	1.0×10^1	- 1,600	3.0×10^3	1.4×10^3	- 114.3
0.8	1.6×10^2	1.0×10^1	- 1,500	3.6×10^3	1.4×10^3	- 157.1
1.0	2.0×10^2	1.0×10^1	- 1,900	3.3×10^3	1.4×10^3	- 135.7

3.4 Conclusion and Recommendation

The results observed in this study have shown that a 1:2 impregnation ratio of ZnCl2 to coconut shell carbon was effective in developing microporous structures with a 0.056 mg/g iodine value. There was an increase in the pore volume distribution on the carbon surface, while the least total coliform and bacteria counts and percentage reductions in the disinfected water samples were obtained with an optimum dose of 0.4 g of GACSC and NaOCl. It should also be noted that GACSC does not possess a sterilizing ability by has been found through this study to significantly reduce microbial load.This research project has revealed that GACSC demonstrated more potential for higher efficiency of water disinfection than NaOCl which is conventionally used. Based on the observations in this present study, it is recommended that granulated $ZnCl_2$-activated coconut shell carbon be used as an alternative to the conventional use of sodium hypochlorite to disinfect drinking water. It is equally recommended that further research be carried out to ascertain the cause(s) of the introduction of some pathogens into the treated water samples.

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